JOURNAL OF AGRICULTURAL RESEARCH

VOL. 41 WASHINGTON, D. C., SEPTEMBER 1, 1930 NO. 5

EFFECT OF CERTAIN CLIMATIC FACTORS ON THE DIAMETER GROWTH OF LONGLEAF PINE IN WESTERN FLORIDA [1]

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INTRODUCTION

Studies of the properties of wood show fluctuating variations that can be attributed only to differences in the structure of the test pieces. Within any one species of tree such variations result from the influences of environmental, and perhaps inherent, factors; hence the departures in properties of the resultant timbers are explained by the effects of these factors on xylem formation. Two methods of approach to the problem present themselves: (1) A study of growth and structure under such conditions that any one environmental factor may be regulated at will; and (2) a statistical analysis of existing stands to determine through correlation the effect of a given factor on growth. The second method has been followed in this investigation.

LITERATURE REVIEW

Data on the inherent factors that exert an influence on diameter increases in woody plants (movements of food materials and water, enzymatic activity, hereditable cyclic activity, and the like) are so meager that statistical methods can not be employed in their analyses. Of the more potent fluctuating environmental factors, precipitation and air temperature are the only ones on which there are records over extended periods of time. Hence the influence of only these two factors can be considered at present.

Precipitation has long been considered as one of the most important factors exerting a controlling influence on growth. Douglass (*6, p. 10*)[3] concluded as follows:

> In a considerable number of cases, but especially in the dry-climate groups' this [correlation between precipitation and diameter growth] has been found to be in the neighborhood of 70 per cent, which is raised substantially by applying a formula to allow for some degree of moisture conservation.

Trees grown in moist climates showed a rhythmic variation in the width of the annual rings of growth that corresponded to the solar activity. The effect of sun spots on tree growth appeared to be through their influence on rainfall. Shreve (*17*) noted a weak qualitative correlation between diameter growth and annual rainfall in Monterey pine, and a small positive relationship in redwood with the December-September precipitation, indicating the beneficial influence of the late winter rainfall on growth in the succeeding season. Pearson

[1] Received for publication June 4, 1930; issued September, 1930.

[2] The writer wishes to thank Eugene W. Gemmer of the United States Forest Service for aid in locating suitable areas for this investigation; B. H. Paul for suggestions given; also Arthur Koehler of the Forest Products Laboratory, under whose general supervision this work was carried on.

[3] Reference is made by number (italic) to Literature Cited, p. 362.

(*14*) decided that the precipitation during April and May had more effect on height growth in western yellow pine than did that during any other portion of the year. Brewster (*2*) was unable to find a definite relation between height growth in western larch and precipitation during the early growth months (April–July). Burns (*3*), working in the Vermont forests says, "There can be no direct correlation between rainfall and diameter growth." Conard (*4*) failed to locate a correlation in Iowa between the rate of growth of oaks and rainfall. Stewart (*18*) showed that the June–July precipitation was more closely related to diameter growth in oak than was the rainfall for the growing season. He studied the annual rings of growth in but one stump, and used records from a meteorological station 25 miles distant. Bogue (*1*), working in Michigan, found that an abnormally small or large precipitation was indicated in the ring of the following year, though Robbins (*15*) could find no evidence of such a delayed effect. Hartig and Weber (*8*) noted that the annual ring in beech formed during a wet year was only 80 per cent of the width of the previous ring, while spruce in the same locality possessed a normal increment.

Attempts to correlate the progression of xylem formation with temperature have not been so successful as have correlations with rainfall. Schwarz (*16*) has recorded a definite relationship between the January-March temperature and ring width in *Pinus sylvestris* Linnaeus. The widest rings were formed in the years having a mean monthly temperature above a 22-year average, and with few 5-day periods in which the average temperature was below the freezing point. Conversely, narrow rings were produced whenever the temperature for the first four calendar months was below the average and showed numerous 5-day periods in which the mean fell below 0° C. Nördlinger (*13*) believed that a warm moist year must favor the development of the ring, and that the preceding winter precipitation played a part. Shreve (*17*), working with Monterey pine, summed all the temperatures above 40° F. and concluded that there was no correlation between the result and tree growth. He stated that "there is rarely justification for attributing irregularities of growth under natural conditions to the fluctuation of a single environmental condition." Robbins (*15*) found that in the red oak group ring width varied inversely as the sum of the mean temperatures for May and June, though a direct correlation with the March–June rainfall was also present. Stewart (*18*) reached no conclusions as to the relation between temperature and growth. Pearson (*14*) noted that height growth in young western yellow pines varied inversely as the temperature and attributed this to the effect on transpiration and water supply.

EXPERIMENTAL FIELD CONDITIONS

Longleaf pine (*Pinus palustris* Miller) was selected for the Forest Products Laboratory study presented here, partly because of its high rating as a structural timber, and partly because the results might be applicable to other experiments under way. The data were obtained on the Choctawhatchee National Forest in western Florida, approximately 50 miles east of Pensacola and about 7 miles from the Gulf of Mexico. The particular trees examined in this investigation

were growing on Section 15, Range 23 West, Township 1 South (Tallahassee Meridian).

This region receives an annual rainfall of about 60 inches. The mean for the early spring months is lower than that for the others, while July and August show a greater precipitation. Most of the rainfall during the vegetative season occurs in heavy showers over small areas. The growing season, as determined by the first and last frosts, is from early February until late November. The weather records of the Camp Pinchot ranger station, 4½ miles to the southwest of the trees studied, were used throughout this investigation.

The portion of the township in which the study was conducted supports a rather open stand of longleaf pine (*Pinus palustris* Miller) with its attendant understory of turkey oak (*Quercus catesbaei* Michaux), post oak (*Q. stellata* Wangenheim), blue-jack oak (*Q. cinerea* Michaux), and palmetto (*Serenoa serrulata* (Michaux) Hooker). The stand is on a poor site. The trees, which are slow in growth, range from 150 to 250 years of age.

This area has an almost imperceptible slope to the east and northeast, and the subterranean water movements are in the same direction. The soil is Norfolk sand of undetermined depth and with a maximum capillary water-holding capacity of 8 per cent. Some of the larger trees had been cut at various times on this forest area prior to 1908, the stand has not been turpentined, and has suffered little from fire since 1914, when it was acquired by the Forest Service. The available records show a possible light fire in 1920, and controlled burning to remove débris in 1924 and in 1926.

Fifty-seven trees, ranging in diameter from 2 to 19 inches at breast height, were selected for study. They were carefully measured and described in the field, and increment cores were taken at a height of 4½ feet above ground from four sides, corresponding to the cardinal compass points. All measurements of ring widths were made under a microscope with the aid of a mechanical stage permitting readings to 0.1 mm. and were made continuously from the outside toward the center. In this way any error in measurement or computation was restricted to the half rings in which it occurred.

RELATION BETWEEN TEMPERATURE AND WIDTH OF RING

The investigations of the effect of temperature on xylem production in longleaf pine in western Florida have led to no positive results. In no case was there a combination of monthly mean temperatures that could be considered as influencing diameter growth. Summations of the temperatures above 40° F. in hour-degree units showed such a close correspondence between the various years that no conclusions could be drawn from them. Shreve (*17, p. 115*) had found previously that the same conditions held for Monterey pine in the "equable maritime climate of Carmel," Calif.

RELATION BETWEEN PRECIPITATION AND WIDTH OF RING

A review of the literature indicates that in many localities precipitation exerts a measurable influence on ring width in trees. Consequently a relationship between these factors was sought. Preliminary investigations indicated that the xylem of longleaf pine in western Florida is produced from February until November, the exact duration

depending upon the earliness or lateness of the seasons. Following Douglass's method of using rainfall from the end of one vegetative period to the end of the next, the precipitation from November 16 to November 15 was compared with the ring produced during that period. No agreement was found. An analysis of conditions in Florida will show that this negative result might have been expected. All the precipitation in this region occurs as rain, which is not stored in the deep, loose soil. In the vicinity of Flagstaff, Ariz., where Douglass worked, the precipitation during the dormant season occurs as snow, which is largely released as available water at the beginning of vegetative activity and is retained for a prolonged period in the heavier soil. In longleaf pine in western Florida, then, no allowance for appreciable storage of winter precipitation should be made.

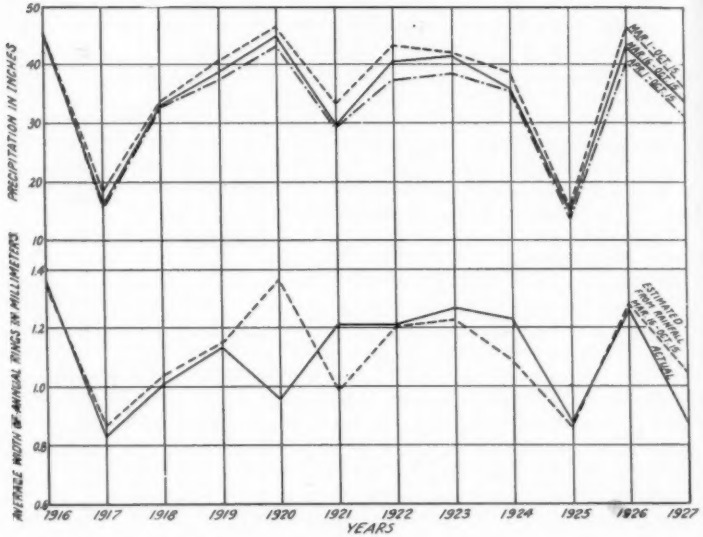

FIGURE 1.—Comparison of seasonal precipitation and average width of annual growth rings by years

Other investigators have indicated the effect on tree growth coming from rainfall that occurs during the early growth months. No such relationship exists in the longleaf pine studied. This may also be attributed to the small storage capacity of the soil and as already stated to the somewhat lower precipitation in this region during the early spring months.

On the other hand, the precipitation occurring during the vegetative season does affect diameter increment, as is evident by reference to Figure 1. The average ring width for each of the 57 trees investigated was derived from four radii. With the exception of the years 1920 and 1921, which are discussed later, the ring width agrees remarkably well with the precipitation. This can be seen best by comparing the curve in Figure 1 showing actual ring width with that show-

ing the width as estimated from the seasonal precipitation. The estimation was based on an average curve in a graph in which ring width was plotted against precipitation during the period of March 16 to October 15 for the years 1916 to 1927, inclusive. The other deviations in the two curves may easily have resulted from varying vegetative periods occasioned by early or late seasons, or from irregular seasonal distribution of the precipitation, although an inspection of the records does not show that the years 1924 and 1927 had exceptionally uneven daily distribution of rain. There is little choice between the period from March 1 to October 15, inclusive, and that beginning 15 or even 30 days later, probably because of the effect of reserve foods, which is discussed later. A qualitative correlation between these values was obtained through the method used by Shreve (*17*). Each year in which the ring width increased or decreased with the precipitation was considered as a positive agreement, and when the two disagreed in direction it was deemed negative. In this way the correlation is 64 per cent using the March 1 to October 15 rainfall, and 73 per cent using that from March 16 to October 15 as well as that from April 1 to October 15. Excluding the years 1920 and 1921, the correlations are 78, 89, and 89 per cent. Two somewhat longer periods, namely, February 16 to October 15, and March 1 to October 31, were discarded because an abundance of rainfall in late February and late October of certain years did not appear to have a corresponding effect on xylem formation. The foregoing data indicate that during the first few weeks of cambial activity sufficient soil moisture, or stored water within the plant, is present under the conditions of relatively low transpiration to allow growth to progress at approximately its maximum rate, and that during the last few weeks growth is so sluggish because of inherent or external factors that it can not be stimulated by an additional supply of water.

The total disagreement of precipitation and xylem production for 1920 and 1921 (fig. 1), regardless of the period for which the precipitation was computed, is not fully explained. Records of the Forest Service show, however, an especially heavy seed crop for longleaf pine in 1920. If this phenomenon were to explain the inconsistencies observed in the curves, there should be some relation between the decrease in ring width and the number of cones borne. The Southern Forest Experiment Station of the United States Forest Service has kindly furnished data as to the number of cones produced on trees of different diameters during a partial seed crop in 1924. Few cones were found on trees up to 8 inches in diameter. The number per tree increased from there on up to a tree diameter of 14 inches, and then decreased. On the basis of this information the trees studied were grouped into diameter classes, and the average ring widths for each group are presented in Figure 2. A comparison of these graphs shows that decreases in radial increments during 1920 as compared with 1919 are roughly in proportion to the potential seed-producing capacity of the various groups.[4] When compared to the expected growth from the precipitation in these two years other facts are evident. The rainfall in 1920 was above that for 1919, hence a wider

[4] It should be observed that while from 1919 to 1920 the growth fell off about the same amount in the trees 5 to 8 inches in diameter as in those 9 to 12 inches in diameter, the decrease on a percentage basis was less in the smaller trees because in general they had wider rings.

ring would be expected. (Fig. 1.) This, coupled with the decrease that actually occurred, accentuates the apparent effect of seed production on ring width.

On the other hand, though the 1921 ring would be expected to be relatively narrow because of moderate precipitation in that year, the rings were actually wider than would be expected from the rainfall, as is indicated in Figure 1. This increase in width of rings may be due to a larger amount of food material stored in the tree or to a stimulus of some kind carried over from the preceding year. Since the seed production did not decrease the width of rings for the following year, two possibilities as to the source of the food material necessary for seed maturation present themselves. Either the food reserves are utilized during the seed year at the expense of ring width, and new reserves are built up later in the season to be used during the following

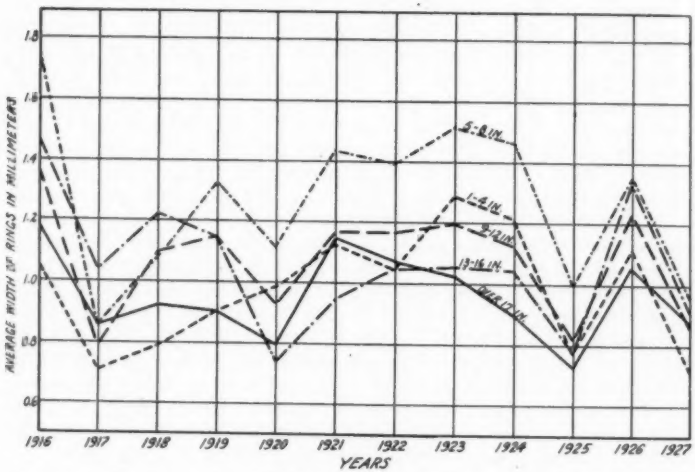

FIGURE 2.—Comparison of average width of annual growth rings in trees of different diameter classes

year, or the reserves of the year preceding fruit production are held in abeyance to be used in xylem production the year after fruiting, and the food materials produced in the current year are diverted largely to seed formation.

Whether the seed year of 1920 actually was the cause of the inconsistencies of ring width in comparison to rainfall in 1920 and 1921 can not definitely be determined, but at present it is the only plausible explanation. Subsequent seed crops, especially in years of abundant rainfall, will be necessary for more conclusive evidence.

EFFECT OF VIGOR OF TREES IN RESPONSE TO RAINFALL

The number of individuals necessary to secure reliable results in any problem to which statistical methods may be applied is always a matter of interest. Douglass (*6*) found that 5 trees of western yellow pine gave almost the same growth, even to small details, as did

15, but that there was less agreement between the oldest pair than among the other three. It should be recalled that most of his trees were of great age. The fact that the older trees, which were probably less vigorous, varied from the mean is also significant.

The preceding discussion has made evident the fact that trees of different diameters vary in response to stimulation. It remains to be

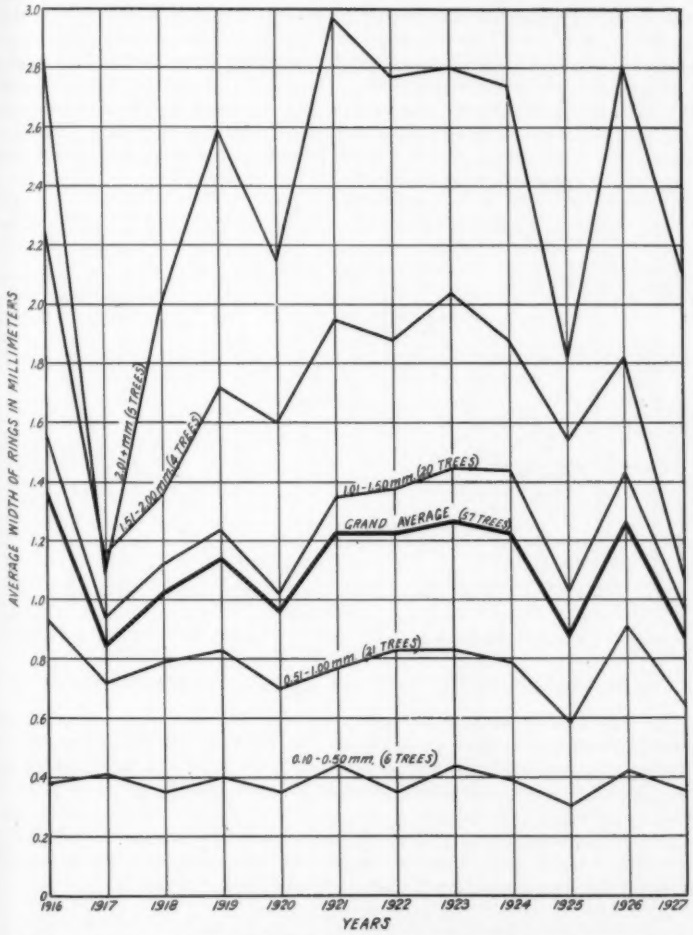

FIGURE 3.—Comparison of average width of annual growth rings by vigor classes as determined by average width of rings

determined whether trees of the different vigor classes show comparable growth responses and whether the results secured from one side of the tree are comparable to those secured from four radii. Investigations of these points were made by comparing curves, and with no attempt to develop numerical comparisons.

Vigor classes were determined from the average width of rings grouped by ½-mm. intervals for 12 years (1916–1927). The mean of each of these arbitrarily selected vigor classes gave a curve that follows with remarkable similarity the general curve for the 57 trees. (Fig. 3.) The departures usually are in amount rather than in direction. The groups with a growth approaching the average show an almost exact duplication of the general curve; the group of slowest growth shows the least similarity in direction and amount of growth. Except for this group of slowest growth, the responses to rainfall were so similar that on a percentage basis they were practically identical in trees of various rates of growth. It is also obvious from Figure 3 that vigorous trees are better visual indicators of the abnormal years than are trees of slow growth, and that narrow rings serve best as indicators to be used in the cross-identification of rings in different trees. Douglass (*6*) also reached similar conclusions.

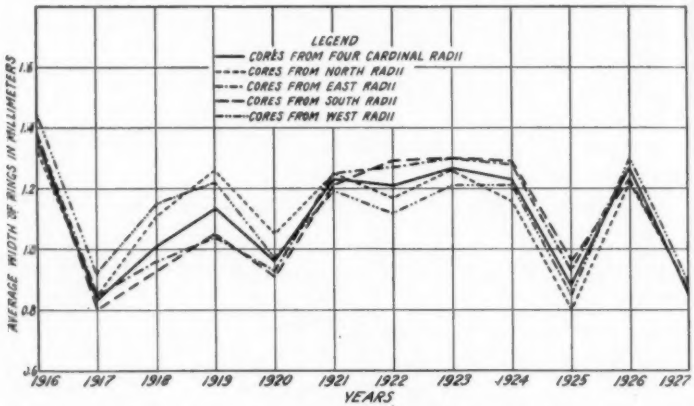

FIGURE 4.—Comparison of average width of growth rings along four individual cardinal radii with the average of four

RELIABILITY OF ONE CORE AS COMPARED TO FOUR

The average growth curve obtained from four cores taken at the cardinal compass points on each of the 57 trees is compared to the average curve for each of the radii in Figure 4. A number of departures of the separate radii from the average for all four may be noted, especially in certain years, although in general the data obtained from any one radius are of about the same magnitude of reliability as the data secured from averages of four cores on different sides when taken from a considerable number of trees.

CARDINAL DIRECTIONS SHOWING GREATEST GROWTH

It is occasionally stated that in north temperate regions straight trees growing on an essentially level site will show an eccentricity in diameter, and that the south and west sides will evince the greater growth. This is attributed to a longer period of activity on these exposures because of greater insolation. Lodewick (*10*) was unable to find evidences of longer cambial activity on the south sides of trees

investigated by him, while Douglass (*6*) reported greatest growth on the northeast side in western yellow pine because the slope of the area on which he worked made soil water first available on that side. The water factor would hardly be operative in the longleaf pine of western Florida where an excess of water in the soil passes downward so readily that lateral movements undoubtedly occur only below the reach of all except the taproot. If such a factor were operative the south and perhaps the west radii should be the longer owing to the coordination of this factor and insolation. The data presented in Figure 4 do not indicate consistently greater increments along any one radius. The cores from each tree were studied separately, and the number of times the total growth for the last decade on each radius was greatest, next greatest, and so on, was recorded. The results for 48 trees, which are given in Table 1, show that the growth along the north and the west radii was larger than along the other two radii. This might presuppose a northwest-southeast eccentricity, though an examination of 12 trees that had recently been cut along a right of way gave no evidence of consistent elongation of any one axis. The decadal growth on each radius permits a similar interpretation in that the north and west sides show a greater total growth.

TABLE 1.—*Relation between cardinal directions and diameter development at breast height in 48 longleaf pines in western Florida*

Radius	Number of times length of each radius ranked—				Average width in mm. of 10-years' growth (1918–1927) on each radius
	First	Second	Third	Fourth	
North	18	13	10	7	12.0
East	10	9	18	11	11.4
South	4	14	12	18	11.2
West	16	12	8	12	11.9

DIAMETER ECCENTRICITY AS RELATED TO CROWN DEVELOPMENT

It is generally supposed that with a restricted horizontal movement of food materials in a tree the diameter increments of that portion of the xylem connected with the larger branches or the greatest number of branches should be the greatest (*11*). Whether or not exposure at breast height directly under the greatest photosynthetic area will show these xylem increments depends upon the straightness of the grain through the intervening bole. Spiral grain, as seen in dead and peeled trees, is frequent in longleaf pine on the Choctawhatchee Forest, though no bark or other external criteria have been found to indicate spiral grain in trees with the bark on. Consequently no information on spiral grain in the trees studied was obtained. The data on the relation between crown development and eccentricity in the bole at breast height have been summarized in Table 2.

TABLE 2.—*Relation between crown and bole development in longleaf pines in western Florida*

Crown development	Number of trees	Radius examined	Bole development—Number of cardinal radii showing growth for 10 years (1918-1927), that is—		
			Least	Medium	Greatest
North least	6	N.	2	0	2
		E.	3	2	0
		S.	1	2	0
		W.	0	2	4
East least	6	N.	0	1	2
		E.	1	0	3
		S.	2	4	0
		W.	3	1	1
South least	3	N.	1	0	1
		E.	2	0	0
		S.	0	2	1
		W.	0	1	1
West least	6	N.	2	0	2
		E.	3	2	0
		S.	1	2	0
		W.	0	2	4
Symmetrical	17	N.	2	4	7
		E.	5	5	3
		S.	4	6	1
		W.	6	2	6

In the trees studied there is no consistent decrease in diameter on the side of least crown development when measurements are made at breast height; in fact, according to the data in Table 2, an increase in diameter was more often the case. In trees supporting a symmetrical crown the north and west sides show a better growth, agreeing with the results obtained when all trees were studied regardless of crown development. The data permit no conjectures as to the reason for this.

CROWN SIZE AND RING WIDTH

An attempt to correlate crown size with diameter increments met with failure. The solid shape of the crown in longleaf pine varies from oval in young trees through a truncated cone to nearly a disk in old trees. By using the cone as the standard, the crown volume of each tree was calculated and compared with the average diameter during the last decade. So little agreement was found, even among similar crown shapes, that it was deemed unnecessary to make more accurate volume computations.

FACTORS AFFECTING SUMMER WOOD PRODUCTION

It has already been shown that the ring width during any one season evinces a close agreement with the precipitation in that period. As a result, it is logical to expect that the rainfall during the time of summer wood formation also affects that portion of the ring. Investigations of longleaf pine during 1927 showed that the heavy-walled part of the ring started during late May or June, the exact time depending upon the season, and continued until the cessation of growth in November. Trials with various beginning and ending

dates on longleaf pine have led to the conclusions that the precipitation from about the middle of June to the middle October has the greatest effect on the production of summer wood, the qualitative correlation in this case being 91 per cent. The curves presented in Figure 5 show that, except for one or two years, there is little choice between the seasons beginning June 1, June 16, or July 1. The disagreement that was noticeable in the total ring width in 1921 is also evident here, but in 1920 the summer wood formation fell off with the rainfall.

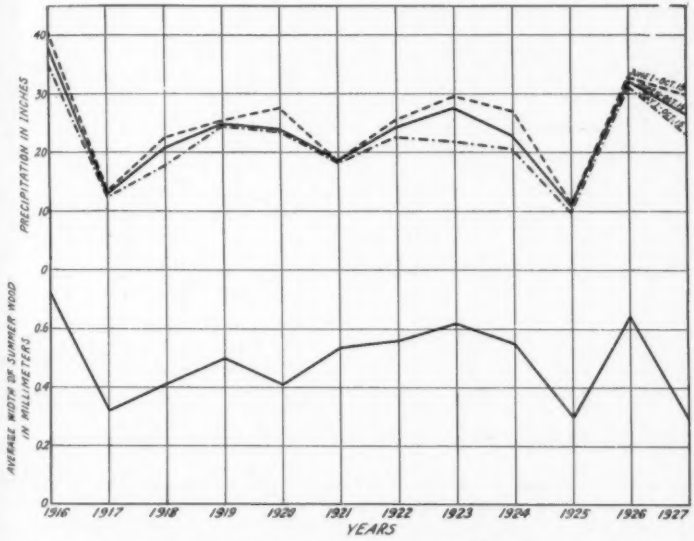

FIGURE 5.—Comparison of seasonal precipitation and average width of summer wood by years

RELIABILITY OF SUMMER WOOD DETERMINATIONS FROM ONE SIDE OF TREE

The reliability of the percentage of summer wood obtained by measurements on one radius as compared with the results from four radii on the same tree was determined by computing the mean and probable error in each measurement. Twenty-five trees were used with the following results:

	Mean summer wood per cent
Four radii	43. 30 ± 0. 246
North radius	44. 07 ± . 490
East radius	43. 07 ± . 499
South radius	43. 21 ± . 480
West radius	42. 85 ± . 485

It is evident that the use of one radius from 25 trees gives results sufficiently accurate for most purposes.

FACTORS AFFECTING SPRING WOOD PRODUCTION

The foregoing discussion has shown that the summer wood is affected by the precipitation of the vegetative period beginning between June 1 and July 1, and, therefore, it is logical to expect that the rainfall during the early portion of the growing season affects the production of spring wood. Figure 6 presents the average spring

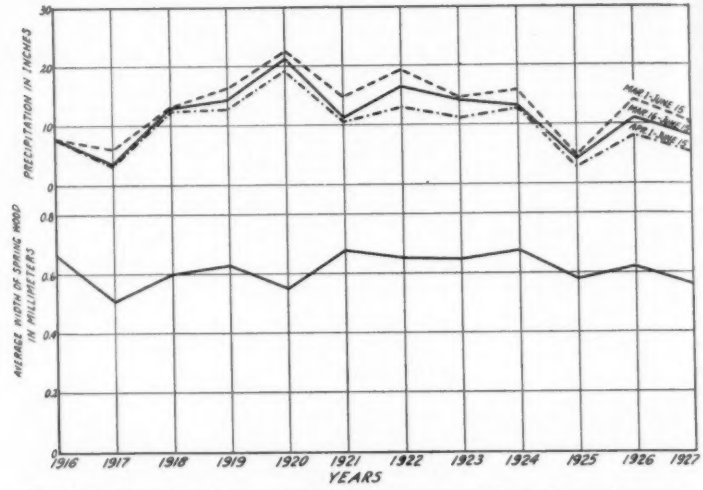

FIGURE 6.—Comparison of seasonal precipitation and average width of spring wood by years

wood width for the 57 trees and the precipitation for three periods during the early months. All three of the periods give fair agreement with spring wood production, though the spring wood did not fluctuate with rainfall to so great a degree as did the summer wood. (Figs. 5 and 6.) The departures are mainly quantitative, the qualitative ones being confined to the years 1920, 1921, 1922, and 1923 for each of the periods, and also for 1924 for the period March 16 to June 15. Apparently other factors also exert an influence on spring wood production.

Since the time of Hartig there has been an hypothesis that the spring wood portion of the ring is formed largely at the expense of stored food reserves (*9*). This seems true enough in the deciduous ring-porous species which, as Lodewick (*10*) has shown, complete the production of the early part of the ring prior to final leaf expansion. There may be some doubt, however, for evergreen species where the old foliage is capable of photosynthetic activity during the period of spring wood formation because the newly elaborated food might be directed to wood production, to foliage development, or to both. De Smidt (*5*) and Myer (*12*) have shown that the ring width is proportional to the volume available for food storage. If the spring wood of any one year is partially dependent upon the stored reserves, a certain amount of spring wood should be formed as long as the rainfall during the early months is sufficient to release the stored reserves

for growth. Figure 7 shows that as the precipitation increases the yearly growth of spring wood formed per unit of water decreases;[5] in fact, except for slight variations, as is also borne out by Figure 6, the spring wood formation is almost a constant, irrespective of rainfall. The summer wood, on the other hand, appears to vary more directly with the rainfall, as is indicated by its more nearly horizontal direction in Figure 7. A combination of the two curves would give an intermediate result for the variation of the total ring width.

Shreve (*17*) utilized similarly derived values in his measurement of the relationship between precipitation and growth, and concluded that there was no relationship between the two because slow-growing trees possessed a higher growth rate per unit of precipitation. An analysis similar to the foregoing would undoubtedly have shown that allowances for the wood formed by reserves should have been made in western yellow pine as well as in longleaf pine.

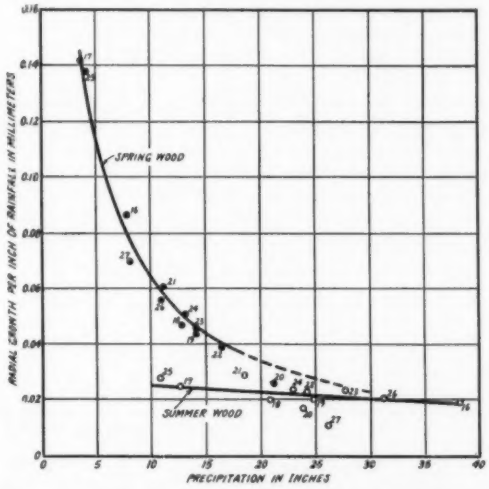

FIGURE 7.—Relation of seasonal growth to seasonal precipitation. The numbers correspond to the years for which the measurements were made

SUMMARY

In an investigation of the relation between certain climatic factors and diameter growth in longleaf pine on the Choctawhatchee National Forest in western Florida no effect of temperature on wood production was found. There was a fairly definite relation between the width of the annual rings and the precipitation from March 16 to October 15 except for the years 1920 and 1921, for which the seed crop of 1920 and its reaction during the following year may have been responsible.

Marked reduction or increase in precipitation was accompanied in most cases by corresponding variations in the ring width.

[5] In Figure 7 the lower extension of the spring wood curve is based on an apparent maximum yearly production of 0.7 mm. spring wood obtained under a water supply equivalent to fully 30 inches of rainfall during spring on an irrigated plot in western Florida.

Vigorous trees were better visual indicators of the effect of climatic factors on wood production than were nonvigorous trees, since the sinuosities of their growth curves were more marked.

Dry years and their accompanying narrow rings were found to be the most reliable for use in the cross identification of rings in different trees.

The data from one radius on 57 trees was about as reliable as the data from four radii on the same trees.

No marked relationship between cardinal directions and diameter growth at breast height was found in the trees investigated.

No relationship between unsymmetrical crown development and eccentric diameter growth was found in the trees investigated.

There appeared to be no correlation between crown size and rate of diameter growth.

The precipitation during the latter part of the growing season (from June 1 to July 1 until October 15) exerted the principal controlling influence on summer wood formation.

The percentage of summer wood as determined from one radius on 25 trees was about as reliable as that from four radii on the same trees.

The precipitation accompanying the period of spring wood formation showed a fair qualitative correlation with the width of the spring wood. Quantitatively this correlation was poorer than that for the summer wood; in fact, over a period of years the width of spring wood was almost constant irrespective of rainfall.

The amount of spring wood formed per unit of rainfall decreased as the precipitation for the season increased, but the amount of summer wood formed was almost directly in proportion to rainfall.

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PARASITES OF THE ORIENTAL PEACH MOTH, LASPEYRESIA MOLESTA BUSCK, IN NORTH AMERICA[1]

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INTRODUCTION

Because of the continued failure to discover a satisfactory control measure for the oriental peach moth (*Laspeyresia molesta* Busck), a pest which is spreading rapidly each year in the more eastern peach-producing areas of the United States and Canada, considerable attention is now being devoted to the insect parasites which attack it.

Complete records of the presence and effectiveness of parasites of this insect have been published thus far for only a few localities. In view of the fact that a search is soon to be made for foreign parasites which might be introduced into this country to aid in combating this insect, it seems advisable at this time to prepare a list of those species which have been recorded to date as parasitic on the pest in North America.

Before parasites from other sources are brought into this country a thorough study should be made throughout the entire range of the host to determine what parasites are already present in each locality, and their abundance and effectiveness. The writer has conducted such a study at Riverton, N. J., for the peach district represented by that locality. The information obtained regarding the parasites in that district is presented here in detail, with a description of some of the methods which proved to be most satisfactory for conducting a survey of oriental peach moth parasites in any locality.

INSECTS RECORDED AS PARASITIC ON THE ORIENTAL PEACH MOTH IN NORTH AMERICA

In connection with biological studies on the oriental peach moth conducted at Riverton and Moorestown, N. J., during four seasons (1925 to 1928, inclusive) the writer found 28 species of primary parasites attacking this host, and 5 species of secondary parasites. By supplementing these records with those obtained from published data and through correspondence,[2] the list of insects recorded as parasitic on the oriental peach moth and presented in Table 1 has been prepared.[3] Information regarding the locality from which each

[1] Received for publication Apr. 2, 1930; issued September, 1930.

[2] In February, 1927, a survey of the more important parasites and predators of the oriental peach moth was compiled by Alvah Peterson and the writer from reports submitted by several cooperators through correspondence and from published data. For the records taken from that survey, from published data, and from correspondence, the writer is indebted to the following workers (who furnished data from the Province and States named in parentheses): W. A. Ross (Ontario, Canada); Philip Garman (Connecticut); D. M. Daniel (New York); L. A. Stearns, B. F. Driggers, and Alvah Peterson (New Jersey); A. B. Champlain, J. R. Stear, S. W. Frost, and J. R. Eyer (Pennsylvania); Philip Garman, H. S. McConnell, and L. A. Stearns (Maryland); L. A. Stearns, W. B. Wood, and E. R. Selkregg (Virginia); and O. I. Snapp (Georgia).

[3] The writer is also indebted to R. A. Cushman and A. B. Gahan, of the Division of Taxonomy and Interrelations of Insects, Bureau of Entomology, and J. M. Aldrich, of the Division of Insects, U. S. National Museum, for the determination of specimens, and particularly to Mr. Cushman for checking the list of species in Table 1.

Journal of Agricultural Research,
Washington, D. C.

Vol. 41, No. 5
Sept. 1, 1930
Key No. K-213

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species has been recorded, the stage of the host which is attacked, and the relation of the parasite to the host is included. The list contains 57 species of primary parasites, 5 of which are questionable, and 8 species of secondary parasites. Ten families of Hymenoptera are represented, including 33 species of Ichneumonidae, 16 of Braconidae, 3 of Eupelmidae, 2 of Pteromalidae, and 1 each of Bethylidae, Eulophidae, Eurytomidae, Perilampidae, Tetrastichidae, and Trichogrammidae. Five species of the dipterous family Tachinidae are also included.

TABLE 1.—*List of insects recorded as parasitic on the oriental peach moth in North America*

PRIMARY PARASITES

Species	Order	Family	Stage of host attacked	Relation to host	Recorded from—
Actia pilipennis Fallen [a]	Diptera	Tachinidae	Larva	Internal	New Jersey.
Aenoplex betulaecola Ashm.[a]	Hymenoptera.	Ichneumonidae	Prepupa [b]	do	Ontario, New Jersey, and Pennsylvania.
Aenoplex carpocapsae Cush.	do	do	do.[b]	do	Pennsylvania.
Allocota thyridopterygis Riley.[c]	do	do	Larva [b]	do	Do.
Angitia sp.[a]	do	do	do	do	New Jersey.
Apanteles harti Vier.[a]	do	Braconidae	do	do	Do.
Apanteles sp.[a]	do	do	do	do	New Jersey, Virginia, and Georgia.
Ascogaster carpocapsae (Vier.)[a]	do	do	Egg	do	Ontario, New Jersey, Pennsylvania, Maryland, and Virginia.
Bassus carpocapsae Cush.[a]	do	do	Larva	do	New Jersey.
Calliephialtes grapholithae (Cress.)[a]	do	Ichneumonidae	Cocoon [d]	External	New Jersey and Pennsylvania.
Calliephialtes n. sp.[a]	do	do	do	do	New Jersey.
Centeterus ineptifrons Gahan.[a]	do	do	do	Internal	New Jersey, Pennsylvania, and Virginia.
Cremastus forbesii Weed [a]	do	do	Larva	do	New Jersey.
Cremastus minor Cush.[a]	do	do	do	do	Ontario and New Jersey.
Cremastus sp.	do	do	do	do	New Jersey.
Cryptus vinctus (Say)	do	do	Cocoon [b]	do	Do.
Diapetimorpha orba (Say)[a]	do	do	Larva [b]	do	Virginia.
Dioctes obliteratus (Cress.)[a]	do	do	do	do	Ontario and New Jersey.
Encyrtaspis semirufus Gahan	do	Eupelmidae	do	do	Georgia.
Ephialtes aequalis (Prov.)[a]	do	Ichneumonidae	Cocoon [b]	do	Ontario, New Jersey, and Virginia.
Epiurus indagator (Cress.)[a]	do	do	Larva or prepupa [b]	External	New Jersey and Maryland.
Epiurus n. sp.	do	do	do.[b]	do	Ontario.
Eubadizon gracilis Prov.[f]	do	Braconidae	Larva	Internal	New Jersey and Virginia.
Eubadizon pleuralis Cress.[f]	do	do	do	do	Ontario and New Jersey.
Eubadizon sp.[a, f]	do	do	do	do	Ontario, Connecticut, New Jersey, and Georgia.
A species of the family Eupelmidae.	do	Eupelmidae	do	External	Maryland.
Glypta phoxopteridis Weed [a]	do	Ichneumonidae	do	Internal	New Jersey.
Glypta rufiscutellaris Cress.[a]	do	do	do	do	Ontario, Connecticut, New York, New Jersey, Pennsylvania, and Maryland.

[a] Bred from *L. molesta* by the writer.
[b] The stage of host attacked is not known, but the probable stage is given.
[c] This species is usually bred from bagworms and tussock moths. Cushman is inclined to doubt that it is parasitic on *L. molestia*.
[d] By cocoon is meant either prepupa or pupa within the cocoon.
[e] Normally parasitic on spider eggs. Cushman doubts the record.
[f] Cushman and Gahan are of the opinion that all these are probably the same and are neither *gracilis* nor *pleuralis* but represent undescribed species.

TABLE 1.—*List of insects recorded as parasitic on the oriental peach moth in North America*—Continued

PRIMARY PARASITES

Species	Order	Family	Stage of host attacked	Relation to host	Recorded from—
Glypta varipes Cress.	Hymenoptera.	Ichneumonidae.	Larva	Internal	Ontario.
Glypta vulgaris Cress.	do	do	do	do	Virginia.
Goniozus sp.	do	Bethylidae	do	External	Do.
Hoplocryptus incertulus Cush. [a] (=incertus Prov.)	do	Ichneumonidae	Cocoon [b]	Internal	New Jersey.
Itoplectis conquisitor (Say) [a]	do	do	do	do	New Jersey, Pennsylvania, and Virginia.
Leskiomima tenera Wied	Diptera	Tachinidae	Larva	do	Maryland.
Leucodesmia nigriventris Gir.	Hymenoptera.	Eulophidae	do	do	Virginia.
Lixophaga plumbea Aldr.	Diptera	Tachinidae	do	do	Do.
Lixophaga variabilis Coq. [a]	do	do	do	do	New Jersey, Maryland, Virginia, and Georgia.
Macrocentrus ancylivora Roh. [a]	Hymenoptera.	Braconidae	do	do	Connecticut, New Jersey, Pennsylvania, Maryland, and Virginia.
Macrocentrus delicatus Cress. [a]	do	do	do	do	New York and New Jersey.
Macrocentrus sp.	do	do	do	do	Ontario, New Jersey, and Virginia.
Meteorus hyphantriae (Riley).	do	do	do	do	New Jersey.
Microbracon gelechiae(Ashm.).	do	do	do	External	New Jersey, Maryland, and Virginia.
Microbracon hebetor (Say) [a]	do	do	do	do	New Jersey.
Microbracon mellitor (Say)	do	do	do	do	Ontario.
Microcryptus sp.	do	Ichneumonidae	Cocoon [b]	Internal	New Jersey.
Nemorilla phycitae LeB.	Diptera	Tachinidae	Larva	do	Maryland.
Phanerotoma tibialis Hald. [a]	Hymenoptera.	Braconidae	Egg	do	New Jersey.
Phygadeuon sp.	do	Ichneumonidae	Larva	do	Do.
Pristomerus ocellatus Cush. [a]	do	do	do	do	Do.
Rogas platypterygis Ashm.	do	Braconidae	do	do	Virginia.
Sagaritis consimilis (Ashm.)	do	Ichneumonidae	do	do	New Jersey.
Sagaritis patsuiketorum Vier.	do	do	do	do	Do.
Spilocryptus sp.	do	do	Cocoon	do	Maryland and Virginia.
Stilbopoides sesiavora Roh.	do	do	Larva	do	New Jersey.
Syntomosphyrum esurus Riley.	do	Tetrastichidae	Pupa	do	Do.
Trichogramma minutum Riley. [a]	do	Trichogrammidae.	Egg	do	Connecticut, New York, New Jersey, Pennsylvania, and Maryland.
Triclistus curvator (Fab.) [a]	do	Ichneumonidae	Pupa	do	New Jersey and Maryland.

SECONDARY PARASITES

Species	Order	Family	Recorded from—
Cerambycobius sp. (probably Eupelmus amicus Gir.).	Hymenopters	Eupelmidae	Virginia.
Dibrachys boucheanus (Ratz.) [a]	do	Pteromalidae	Ontario, New Jersey, Pennsylvania, Maryland, and Virginia.
Dibrachys (Tritneptis) hemerocampae Gir. [a]	do	do	New Jersey.
Eurytoma sp.	do	Eurytomidae	Maryland.
Gelis sp. [a]	do	Ichneumonidae	New Jersey.
Hemiteles tenellus (Say) [a]	do	do	Do.
Hemiteles sp.	do	do	Do.
Perilampus sp. [a]	do	Perilampidae	Do.

[a] Bred from *L. molesta* by the writer.
[b] The stage of host attacked is not known, but the probable stage is given.
[c] Normally a parasite of meal moths.

Although the oriental peach moth has so many insect parasites, very few seem to be of much importance. Those of chief importance are *Macrocentrus ancylivora* (fig. 1) and *Glypta rufiscutellaris* (fig. 2), both of which attack the larval stage, and *Trichogramma minutum*, which attacks the egg.

Macrocentrus ancylivora particularly, and *Glypta rufiscutellaris* to a certain extent, are aiding considerably in reducing the annual infestation in those sections of New Jersey where they have become established. The work of these two species is confined largely to the earlier broods of host larvae that feed in peach twigs. By destroying large numbers of larvae of the first and second broods early in the season they bring about a considerable reduction in the number of individuals produced in the later broods, most of which feed in the fruit. As pointed out by Stearns,[4] these two species are apparently limited in their distribution by environmental factors. Thus far, both species have not seemed able to become successfully established and effective in the same locality.

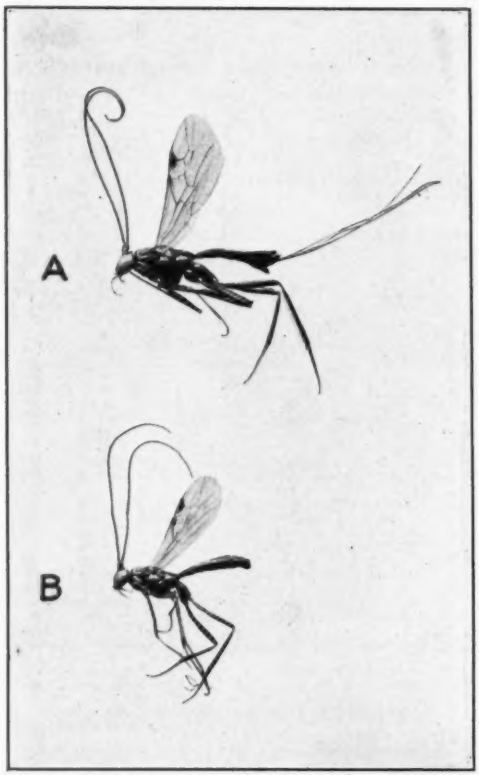

FIGURE 1.—Adults of *Macrocentrus ancylivora*: A, Female, B, male. × 5

The egg parasite *Trichogramma minutum* has been found to attack large numbers of oriental peach-moth eggs placed in peach orchards at Riverton, N. J. Garman [5] and McConnell [6] both record this species as attacking eggs of the oriental peach moth in Maryland. Garman also records it from Connecticut,[7] and Stear records it from Pennsylvania.[8]

Of the remaining species, only a few have been recorded as fairly abundant in some localities. The majority are merely occasional records, usually of only a few individuals.

[4] STEARNS, L. A. THE LARVAL PARASITES OF THE ORIENTAL PEACH MOTH (LASPEYRESIA MOLESTA BUSCK) WITH SPECIAL REFERENCE TO THE BIOLOGY OF MACROCENTRUS ANCYLIVORA ROHWER. N. J. Agr. Expt. Sta. Bul. 420, 24 p., illus. 1928.

[5] GARMAN, P. THE ORIENTAL PEACH PEST (LASPEYRESIA MOLESTA BUSCK), A DANGEROUS NEW FRUIT INSECT OF MARYLAND. Md. Agr. Expt. Sta. Bul. 209, 16 p., illus. 1917.

[6] MCCONNELL, H. S. THE ORIENTAL FRUIT MOTH. Md. Agr. Expt. Sta. Bul. 298: 179-180. 1928.

[7] GARMAN, P. WORK WITH THE ORIENTAL PEACH MOTH IN 1926. Conn. Agr. Expt. Sta. Bul. 285: 234-239. 1927.

[8] STEAR, J. R. THE ORIENTAL FRUIT MOTH IN PENNSYLVANIA. Penn. Dept. Agr. Bul. v. 12, no. 8, Gen. Bul. 477, 13 p., illus. 1929.

PARASITISM OF THE ORIENTAL PEACH MOTH IN THE VICINITY OF RIVERTON, N. J.

METHODS OF OBTAINING RECORDS

The presence of parasites which attack and develop within the egg of the host was determined by placing oriental peach-moth eggs, which had been deposited on the leaves of peach or pear twigs in the insectary, in peach orchards. The twigs, in small bottles of water, were placed on posts in such a position as to be in the foliage of the peach trees. After being exposed for 24 hours or longer, the eggs were taken to the insectary and examined to see if they were parasitized.

Most of the information regarding the species of parasites, which attack or develop within the larva of the oriental peach moth previous to the time the cocoon is spun, was obtained by collecting peach twigs infested with the larvae. Collections were made from several orchards during the four consecutive seasons from 1925 to 1928, inclusive, usually once a week in each orchard from the time the first infested twig was found in the spring until twigs containing larvae could no longer be found in early fall. The collecting was usually continued for a definite number of minutes in each orchard, and the twigs were taken to the insectary immediately. During the first three seasons all the larvae were removed from the twigs, sorted into various size.

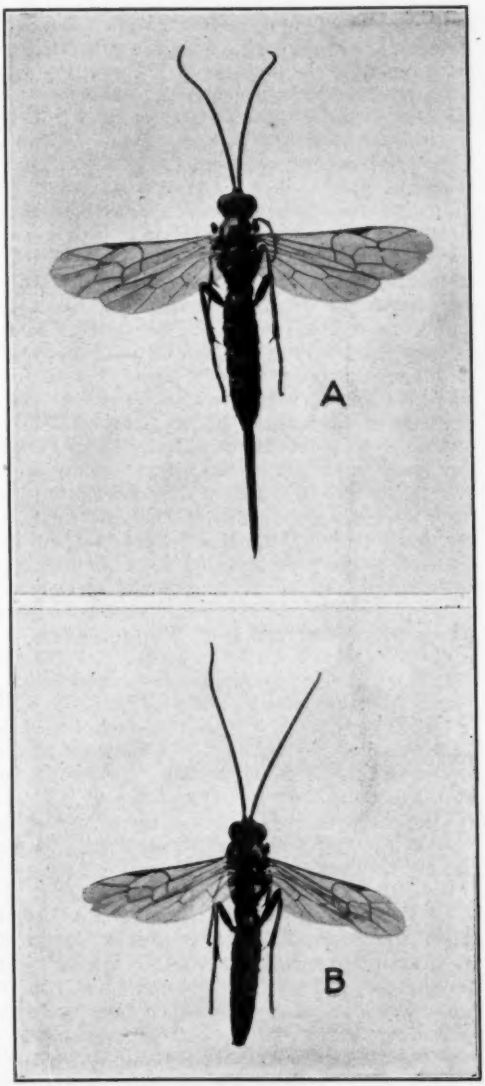

FIGURE 2.—Adults of *Glypta rufiscutellaris*: A, Female, B, male. × 4

according to their length in millimeters, and placed on fruit (peaches or apples) in jelly glasses covered with cheesecloth held in place by rubber bands. They continued their development in the fruit, and then spun cocoons in strips of corrugated straw paper, which were placed in the glasses. The cocoons were removed daily and placed in individual homeopathic vials in which the adult moths or parasites later emerged.

It was found that in rearing the larvae in the insectary after having removed them from the twigs a high mortality occurred, especially in the case of young larvae. Many larvae were injured while being removed from the twigs and died before they could eat their way into the fruit upon which they were placed. Preliminary trials in 1927 indicated that lower mortality resulted if the larvae were not removed from the twigs but were allowed to crawl out at will and go to the fruit. Consequently, throughout 1928 the larvae were removed from only one-half of the twigs taken at each collection. The leaves were trimmed from the remaining twigs, which were shortened to the 3 or 4 inches that contained the larvae. These stems were then placed in 6 by 8 inch glass jars about half full of apples. The more mature larvae, which were able to complete their development within the small portion of twig, crawled out when full grown and spun cocoons in strips of corrugated paper placed in the jars. Larvae which were unable to complete their growth before the twigs dried out abandoned the twigs and completed their feeding in the apples. These also eventually spun cocoons in the corrugated-paper strips. The cocoons were removed about once a week and placed in individual vials until the moths or parasites emerged. Although the same number of infested twigs was handled by each method throughout the entire season, only 1,030 adults (moths and parasites) emerged from the larvae which were removed from the twigs, as compared with 1,433 adults which emerged from the larvae that were allowed to come out of the twigs at will. In other words, 39.1 per cent more individuals completed their development successfully when the larvae were handled by the second method. By being able to rear a larger number of larvae to maturity, a more accurate record can be obtained of the percentage of parasitism occurring in the field. In addition, considerable time is saved because it is not necessary to remove larvae from the twigs.

Additional records of parasites of the larvae were obtained by collecting infested peach and quince fruit in the summer and fall and rearing the larvae to maturity.

Records of the parasites which attack the oriental peach moth while it is a prepupa or a pupa within the cocoon were obtained by collecting cocoons that were spun under the bark of peach and quince trees and rearing the insects to maturity in the insectary. Occasionally larval parasites also emerged from these cocoons, when the host larva had been parasitized before the cocoon was formed. On several occasions strips of corrugated paper containing prepupae or pupae in cocoons were tacked to the trunks or branches of peach trees for several days, but no parasitism occurred.

PARASITISM OF EGGS

Trichogramma minutum was the only parasite found that attacks and develops entirely within the egg of the host. This parasite was present at various times throughout each growing season, and on some occasions 50 per cent or more of the host eggs that were placed on posts in peach orchards were parasitized. Information is not yet available concerning the percentage of parasitism by this species in eggs deposited normally by the oriental peach moth in the orchards near Riverton.

Two species of parasites, *Ascogaster carpocapsae* and *Phanerotoma tibialis*, oviposit in the egg of the host but complete their development within the larva. These two species are treated as larval parasites.

PARASITISM OF LARVAE FEEDING IN PEACH TWIGS

Relative Abundance and Importance of the Various Species

In the four orchards from which twigs were collected during the seasons of 1925 to 1928, inclusive, 14 species of parasites were found which attack or develop within the larva of the oriental peach moth while the larva is feeding in peach twigs. Table 2 shows the species present each season and the relative importance of each. For the four seasons, 91 per cent of all the larval parasites present were of the species *Macrocentrus ancylivora*. *Cremastus minor* accounted for about 3 of the remaining 9 per cent, *M. delicatus* for between 2 and 3 per cent, *C. forbesii* and *Glypta rufiscutellaris* for about 1 per cent each, and all the other species together for only about 1 per cent. *Dibrachys* (*Tritneptis*) *hemerocampae* is a secondary parasite bred from a cocoon of *M. ancylivora*. *Perilampus* sp. also is a secondary parasite. The table shows the percentage of oriental peach-moth larvae that were parasitized by each species during each season and for the four seasons combined. Parasitism by *M. ancylivora* alone ranged from 41.6 per cent in 1928 to as high as 55.3 per cent in 1926, with an average of 48.23 per cent for the four seasons. The percentage of parasitism by no other single species exceeded 2.69 per cent in any given season, and the highest average parasitism over the 4-year period by any species other then *M. ancylivora* was 1.65 per cent by *C. minor*.

Macrocentrus ancylivora is the only larval parasite of importance in the Riverton district, as is clearly indicated in Table 2.

TABLE 2.—*Relative abundance and importance of each species of larval parasite of the oriental peach moth present in peach orchards at Riverton, N. J., during the seasons of 1925 to 1928, inclusive, as determined by collections of twigs from orchards 2 to 6 years old* [a]

Species	1925			1926			1927			1928			4 seasons (1925 to 1928)		
	Number of individuals emerged	Percentage of larvae parasitized	Percentage of all parasites	Number of individuals emerged	Percentage of larvae parasitized	Percentage of all parasites	Number of individuals emerged	Percentage of larvae parasitized	Percentage of all parasites	Number of individuals emerged	Percentage of larvae parasitized	Percentage of all parasites	Number of individuals emerged	Percentage of larvae parasitized	Percentage of all parasites
Macrocentrus ancylivora Roh	437	49.27	91.6	532	55.30	91.0	1,003	52.79	89.0	1,027	41.60	93.3	2,999	48.23	91.2
Cremastus minor Cush	18	2.03	3.8				47	2.48	4.2	38	1.54	3.4	103	1.65	3.1
Macrocentrus delicatus Cress				9	.94	1.5	51	2.69	4.5	25	1.01	2.3	85	1.36	2.6
Cremastus forbesii Weed	17	1.92	3.6	25	2.60	4.3	1	.05	.1	2	.08	.2	45	.72	1.3
Glypta rufiscutellaris Cress	3	.34	.6	17	1.77	2.9	5	.26	.4	4	.16	.4	29	.46	.9
Ascogaster carpocapsae (Vier.)	1	.11	.2				11	.58	.9	3	.12	.3	15	.24	.5
Lixophaga variabilis Coq				2	.21	.3	1	.05	.1				3	.05	.4
Pristomerus ocellatus Cush							3	.16	.3				3	.05	
Apanteles harti Vier							2	.11	.2				2	.03	
Bassus carpocapsae Cush										1	.04	.1	1	.02	
Phanerotoma tibialis Hald	1	.11	.2										1	.02	
Diodes obliteratus (Cress.)							1	.05	.1				1	.02	
Dibrachys (Tritneptis) hemerocampae Gir. [b]							1	.05	.1				1	.02	
Perilampus sp. [b]							1	.05	.1				1	.02	
Parasitism of larvae by all species	477	53.78	100.0	585	60.81	100.0	1,127	59.32	100.0	1,100	44.55	100.0	3,289	52.89	100.0
Total oriental peach moths	410			377			773			1,369			2,929		

[a] The collections in 1925 were from 2 orchards, in 1926 from 3 orchards, and in 1927 and 1928 from 4 orchards.
[b] Secondary parasite.

SEASONAL PARASITISM

The total parasitism of larvae for each season by all species combined and the average parasitism for the four seasons are also given in Table 2. The seasonal parasitism was highest in 1926, when 60.81 per cent of the larvae collected in twigs were parasitized. The lowest parasitism occurred in 1928, when only 44.55 per cent were parasitized. An average of 52.89 per cent of all the larvae collected in peach twigs during the entire 4-year period was killed by various species of larval parasites.

In Table 3 the collections from the several orchards each week during a given season have been combined. This table gives the number of individuals which emerged, including both moths and parasites, the total percentage of parasitism by all species, the percentage of parasitism by *Macrocentrus ancylivora* alone, and the percentage by all other species combined, as they occurred in the weekly collections. In the collections made June 23 and 24, 1925, and July 15, 1926, as high as 95 per cent of the larvae were parasitized. The maximum parasitism in weekly collections in 1927 was considerably lower, 68 per cent, and occurred in the collections of

June 7 to 10 and of July 5 to 7. In the collection of July 3 to 5, 1928, there was a maximum parasitism of 72.1 per cent. In each instance indicated above, the percentage of parasitism by *M. ancylivora* alone also reached the maximum. The figures given in Table 3 further indicate the importance of *M. ancylivora* in the Riverton district.

TABLE 3.—*Percentage of parasitism in individuals which emerged from weekly collections of peach twigs infested with larvae of the oriental peach moth at Riverton, N. J.*

Collection dates	Number of individuals emerged	Percentage of larvae parasitized	Percentage of larvae parasitized by—		Collection dates	Number of individuals emerged	Percentage of larvae parasitized	Percentage of larvae parasitized by—	
			M. ancylivora	All other species				M. ancylivora	All other species
1925					1927				
May 18–21	58	67.2	67.2	.....	May 11–13	7	57.1	42.8	14.3
May 26, 27	148	66.9	64.2	2.7	May 17–19	28	28.6	28.6	.....
June 2, 3	130	36.9	36.9	.....	May 24–26	153	59.5	59.5	.....
June 9, 10	92	13.0	13.0	.....	May 31–June 2	300	63.3	63.0	0.3
June 16, 17	98	24.5	23.5	1.0	June 7–10	399	68.2	64.4	3.8
June 23, 24	109	95.4	92.7	2.7	June 14–16	284	59.1	53.5	5.6
June 30, July 1	96	93.8	71.9	21.9	June 21–23	222	54.0	44.6	9.4
July 7, 8	41	73.2	58.6	14.6	June 28–30	218	52.3	28.0	24.3
July 14, 15	13	46.2	38.5	7.7	July 5–7	152	68.4	63.2	5.2
July 21, 22	6	33.3	33.3	.....	July 12–14	89	51.7	44.9	6.8
July 27, 28	6	50.0	33.3	16.7	July 19–21	7	42.9	28.6	14.3
Aug. 2–6	8	50.0	37.5	12.5	July 26–28	10	10.0	10.0	.....
Aug. 11, 12	14	21.4	14.3	7.1	Aug. 2–4	20	25.0	15.0	10.0
Aug. 18	10	20.0	20.0	.....	Aug. 9–12	10	10.0	10.0	.....
Aug. 25, 26	21	33.3	28.7	4.7	Aug. 17–19	1	.....	.....	.....
Sept. 1	15	6.7	6.7	.....	1928				
Sept. 8	17	5.9	5.9	.....	May 16–19	32	6.3	6.3	.....
Sept. 15	4	50.0	50.0	.....	May 22–24	268	45.9	45.9	.....
Sept. 22	1	.....	.....	.....	May 29–June 1	453	50.1	49.9	.2
1926					June 5–8	622	42.3	39.4	2.9
May 17–19	1	.....	.....	.....	June 12–15	276	29.7	25.7	4.0
May 25, 26	34	47.1	47.1	.....	June 19, 20	168	20.2	17.9	2.3
June 2, 3	87	40.2	40.2	.....	June 26–28	173	40.5	29.5	11.0
June 8, 9	60	33.3	31.7	1.6	July 3–5	237	72.1	67.9	4.2
June 16, 17	60	11.7	11.7	.....	July 10–13	149	69.1	64.4	4.7
June 23	64	9.4	7.8	1.6	July 17, 18	41	43.9	41.5	2.4
June 30	118	37.3	35.6	1.7	July 24, 25	36	11.1	5.6	5.5
July 6, 7	157	88.5	83.4	5.1	July 31–Aug. 2	2	50.0	50.0	.....
July 15	107	95.3	84.1	11.2	Aug. 7–10	7	14.3	14.3	.....
July 21	122	86.9	76.2	10.7	Aug. 14, 15	1	100.0	100.0	.....
July 28	74	58.1	45.9	12.2	Aug. 21	4	.....	.....	.....
Aug. 4	66	90.9	80.3	10.6					
Aug. 11	3	66.6	66.6	.....					
Aug. 18	5	60.0	60.0	.....					
Aug. 25	4	50.0	50.0	.....					

With the exception of 1926 (when only one individual emerged from the first collection) parasitism varying from 6.3 to 67.2 per cent occurred in the first collection each season. It is thus apparent that the parasites emerge early enough in spring to attack the first larvae which enter the twigs. This was checked in the case of *Macrocentrus ancylivora* in the spring of 1928. It was found that these parasites began to emerge on May 11 after having hibernated in host larvae kept outdoors throughout the winter in a wire-screen cage hung on the south side of a pole, a method similar to that used by Peterson and Haeussler [9] to determine the spring-brood emergence of oriental peach moths. This was five days earlier than the first oriental peach-moth larvae were found in twigs in the orchard (May 16).

[9] PETERSON, A., and HAEUSSLER, G. J. DETERMINATION OF THE SPRING-BROOD EMERGENCE OF ORIENTAL PEACH MOTHS AND CODLING MOTHS BY VARIOUS METHODS. Jour. Agr. Research 37: 399–417, illus. 1928.

The most abundant parasitism occurred each season when larvae were most abundant in the twigs, as shown in Figure 3. The bars of this chart also indicate the number of moths, of *Macrocentrus ancylivora*, and of all other species of parasites combined which emerged from infested peach twigs collected weekly each season. The cross-hatched areas indicate the number of oriental peach moths,

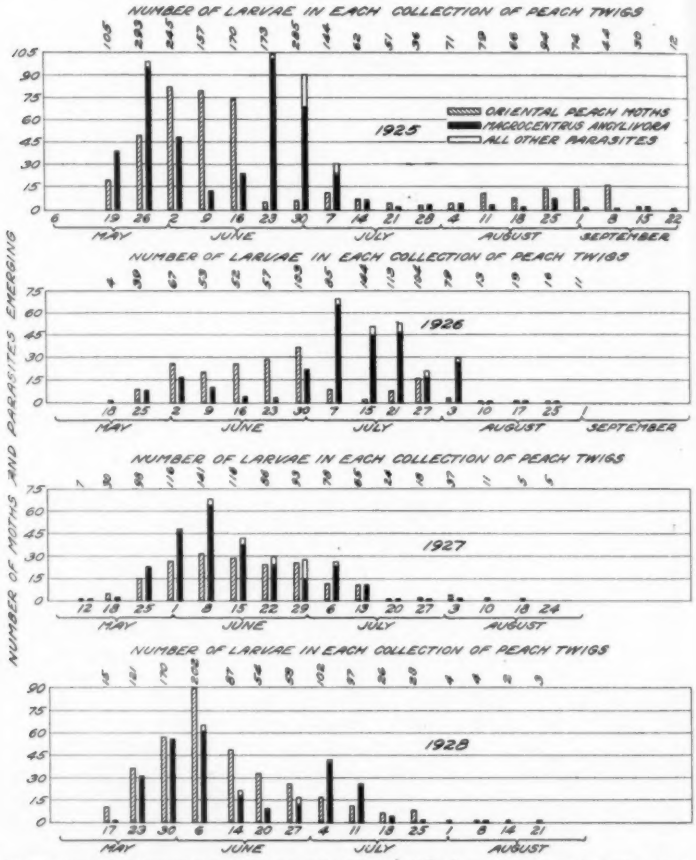

FIGURE 3.—Parasitism of oriental peach-moth larvae feeding in peach twigs at Riverton, N. J., 1925 to 1928, inclusive, as indicated by the number of individuals which emerged from infested twigs collected weekly in 15-minute periods

the solid black areas the number of *M. ancylivora*, and the white outlined areas the total number of parasites of all other species combined. The number of larvae taken in each collection is also given, and, as each collection was made during a 15-minute period, the figures indicate the relative intensity of infestation on each date of collection.

In 1925 a large population of first-brood host larvae was present in the twigs. These larvae were heavily parasitized in May, thus building up a strong population of parasites, chiefly *Macrocentrus ancylivora*, which in turn severely attacked the large second brood of host larvae. The heavy parasitism during the latter part of June and early July caused a decided decrease in twig infestation after the collection of July 7, but larvae were present in the twigs throughout August and until the collection of September 22. Parasitism after the second brood was comparatively low. There were five broods of host larvae at Riverton in 1925.

Host larvae of the first brood were not very abundant in the twigs in 1926, and parasitism of these larvae was low. Host larvae of the second brood were abundant in twigs throughout July and were heavily parasitized. Although twigs suitable for infestation were present in the orchards well into September, very few twigs were attacked after the collection of August 3. Only four broods of host larvae occurred at Riverton in 1926.

In 1927 the host larvae were abundant in the twigs from the collection of May 25 to the collection of July 13, and in every collection made during this period 50 per cent or more of the larvae were parasitized. This continuously heavy parasitism reduced the host population to such an extent that very few larvae were present in twigs after the middle of July. Four broods of host larvae occurred this season.

The situation in 1928 was, in some respects, similar to that in 1925. Larvae of the first brood were the most abundant and were heavily parasitized. Parasitism of larvae of the greatly reduced second brood was very severe early in July, and few host larvae were found in twigs after July 25, although a supply of twigs suitable for infestation was available until about August 10. There were five broods of host larvae in 1928.

PARASITISM OF LARVAE FEEDING IN FRUIT

Parasitism as high as 28 per cent by *Macrocentrus ancylivora* and 7 per cent by *M. delicatus* has been found in larvae feeding in peaches during the months of June, July, and August at Riverton. It is possible that many of these larvae were parasitized while feeding in twigs and that they abandoned the twigs later to complete their feeding in the fruit. From a large number of larvae cut from peaches from late August to early November in 1926, a total of 673 oriental peach moths and 10 *M. ancylivora* emerged the following spring. This was only 1.46 per cent parasitism, all by one species of parasite, in wintering larvae. These larvae probably fed almost entirely in the fruit.

From larvae collected in the fall of 1928 while feeding in quince fruit, five species of primary parasites and one secondary parasite emerged, as shown in Table 4. Of the 949 individuals which emerged, 40 were parasites, a parasitism of 4.21 per cent in larvae feeding in quince fruit at harvest time. *Phanerotoma tibialis*, which oviposits in the egg of the host and develops in the larva, was the most abundant species. The 24 individuals of this species and one of the two *Macrocentrus ancylivora* hibernated in the larvae of the host and did not emerge until the following spring. The other *M. ancylivora* and all the individuals of the remaining species emerged in the fall

of 1928. Of the 40 parasites which emerged from this material, 10 were *Glypta rufiscutellaris*, whereas there were only 4 individuals of this species among 1,100 parasites obtained from larvae feeding in peach twigs collected earlier the same season. (Table 2.)

TABLE 4.—*Parasitism of oriental peach moth larvae feeding in quince fruit at Riverton, N. J., 1928*

Species	Number of individuals emerged		
	Transforming	Wintering	Total
Phanerotoma tibialis Hald.		24	24
Glypta rufiscutellaris Cress.	10		10
Macrocentrus ancylivora Roh.	1	1	2
Calliephialtes grapholithae (Cress.)	2		2
Pristomerus ocellatus Cush.	1		1
Hemiteles tenellus (Say) [a]	1		1
Total parasites emerged	15	25	40
Oriental peach moths emerged	31	878	909
Total individuals emerged	46	903	949
Percentage of parasitism			4.21

[a] Secondary parasite.

PARASITISM AND MORTALITY OF ORIENTAL PEACH MOTHS IN COCOONS COLLECTED FROM UNDER QUINCE BARK

From a number of oriental peach moth cocoons collected under quince bark in the fall of 1928, only 74 individuals emerged, 15 of which, or 20.27 per cent, were parasites. (Table 5.) Eleven of the parasites were *Aenoplex betulaecola*, which attacks the host in the cocoon, probably when it is a prepupa. One of these emerged in the fall of 1928, and 10 hibernated. All the other species represented attack the host while in the cocoon, with the exception of *Pristomerus ocellatus*, a parasite of the larva.

TABLE 5.—*Parasitism and mortality of oriental peach moths in cocoons collected under quince bark at Riverton, N. J., 1928*

Species	Number of individuals emerged		
	Transforming	Wintering	Total
Aenoplex betulaecola Ashm.	1	10	11
Centeterus ineptifrons Gahan	1		1
Ephialtes aequalis (Prov.)		1	1
Hoplocryptus incertulus Cush.		1	1
Pristomerus ocellatus Cush.		1	1
Total parasites emerged	2	13	15
Oriental peach moths emerged	2	57	59
Total individuals emerged	4	70	74
Percentage of parasitism			20.27

Number of oriental peach-moth larvae dead	162
Number of oriental peach-moth pupae dead	15
Total individuals dead	177
Percentage of mortality	70.52

As shown in Table 5, 70.52 per cent mortality occurred in the larvae and pupae in the cocoons collected under quince bark. Most of these were apparently killed by a fungous disease which is common in oriental peach-moth larvae that hibernate in cocoons spun under bark.

SUMMARY

Fifty-seven species of primary parasites attacking the oriental peach moth in North America have been recorded thus far. Those of chief importance are the larval parasites *Macrocentrus ancylivora* and *Glypta rufiscutellaris*, and the egg parasite *Trichogramma minutum*. Eight species of secondary parasites have also been found.

Records of the parasites of larvae occurring in the vicinity of Riverton, N. J., were obtained by making weekly collections throughout the season of peach twigs infested by the host larvae, and rearing these larvae to maturity. By trimming off the leaves and placing the portion of stems containing the larvae on apples in glass jars, 39.1 per cent more individuals were able to complete their development than when each larva was removed from the twig and placed on fruit.

Trichogramma minutum was the only parasite found at Riverton which develops entirely within the egg of the host. *Ascogaster carpocapsae* and *Phanerotoma tibialis* oviposit in the egg of the host but complete their development within the larva.

Fourteen species of parasites of larvae were found in larvae feeding in peach twigs at Riverton during the seasons of 1925 to 1928, inclusive. *Macrocentrus ancylivora* is the only important parasite of larvae in the Riverton district. During the 4-year period, 91 per cent of all the parasites of larvae present were *M. ancylivora*, and parasitism by this species alone averaged 48.23 per cent.

The seasonal parasitism by all species of parasites combined varied from 60.81 per cent in 1926 to 44.55 per cent in 1928, with an average of 52.89 per cent for the four seasons.

Parasitism as high as 95 per cent occurred in weekly collections of larvae in 1925 and 1926.

Macrocentrus ancylivora emerges early enough in the spring to attack the first host larvae that feed in peach twigs.

The most abundant parasitism occurred each season when host larvae were most abundant in twigs. Parasitism of larvae of the first and second broods caused a decided decrease in the number of twigs infested each season.

Parasitism of 28 per cent by *Macrocentrus ancylivora* and 7 per cent by *M. delicatus* occurred in larvae feeding in peach fruit in the early part of the season, but less than 2 per cent parasitism was found in wintering larvae collected while feeding in peaches in the fall.

About 4 per cent of the larvae collected while feeding in quince fruit at harvest time were parasitized, and *Phanerotoma tibialis* was the most abundant parasite. Parasitism by *Glypta rufiscutellaris* was much more abundant in these larvae than in larvae feeding in twigs.

Aenoplex betulaecola was the most common parasite of the stages within the cocoon. High mortality, due to disease, occurred in oriental peach-moth larvae hibernating in cocoons spun under quince bark.

SHORT BRANCH, ANOTHER CHARACTER OF COTTON SHOWING MONOHYBRID INHERITANCE [1]

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INTRODUCTION

Satisfactory evidence of monohybrid or simple Mendelian inheritance has been adduced for relatively few characters of the cotton plant. The following list of pairs of allelomorphs comprises the characters known to the writer for which there has been published fairly conclusive proof of monohybrid segregation in certain crosses.

Plant "crinkled dwarf"; plant normal (*11*).[2]
Leaf narrow-lobed ("okra"); leaf broad-lobed (*4, 8, 20, 22, 23, 26, 27, 28, 32*).
Leaf red; leaf green (*4, 23, 26, 31, 32*).
Corolla whitish; corolla yellow (*1, 3, 7, 8, 12, 15, 17, 20, 21, 23, 24, 26*).
Petal spot absent or weak; petal spot intense (*1, 10, 13, 17, 18, 21, 24, 26*).
Pollen cream-colored; pollen yellow (*1, 2, 4, 14, 17, 26*).
Seeds fuzzy; seeds naked or nearly so (*1, 4, 7, 8, 17, 19, 21, 26, 29, 30*).
Lint sparse; lint abundant [3] (*9, 20, 21, 29, 30*).

A few other characters, in regard to which the evidence for the existence of a single pair of allelomorphs seems less conclusive or is conflicting, are: Color of the leaf pulvinus (*1, 17, 21*), color of the unripe boll (*21*), and color of the fuzz on the seed (*4, 21*).

Several of the characters in the first list have shown clear-cut monohybrid inheritance in crosses between nearly related forms but a more complicated type of inheritance in interspecific hybrids. A good example is spotless or weak-spotted petal as contrasted with full-spotted petal. When a full-spotted and a weak-spotted form of the same species were crossed, all plants of the F_2 population could readily be referred to one or the other group, and the departure of the respective numbers from a 3:1 ratio was insignificant (*18*). On the other hand, when the cross was between different species, e. g., full-spotted Egyptian or sea-island cotton (*Gossypium barbadense*) and spotless upland cotton (*G. hirsutum*), F_2 showed intergradation between the two classes, and separation of them was more or less arbitrary (*13, 17*).

An explanation of this difference in the behavior of crosses between related and unrelated forms has been offered by Harland (*14, p. 399*).

* * * species hybrids in cottons differ from varietal hybrids in the extent to which differences occur in modifying factors. In a species hybrid each main gene is apparently accompanied by a group of modifiers which have the effect of diluting the character in steps down to the recessive, and in certain cases may obscure entirely the distinction between dominant and recessive.

The argument is restated in another paper by the same author (*16*), as follows:

[1] Received for publication May 1, 1930; issued September, 1930.
[2] Reference is made by number (italic) to Literature Cited, p. 386.
[3] The only well-supported instance of definite alternative inheritance of sparse lint as contrasted with abundant lint appears to be the one described by Thadani (*29, 30*). In this case there seemed to be close linkage with naked seed as contrasted with fuzzy seed, and it may be that only different expressions of a single gene are involved.

In varietal crosses there is segregation of the main gene alone, whereas in the interspecific cross there is segregation of modifiers which may partially, or in some cases entirely, obscure the distinction between dominant and recessive. Thus, there is no real difference in the mechanism of inheritance in interspecific from that which exists in varietal crosses.

The difference between crosses within a species and interspecific hybrids may extend to a reversal of dominance. Thus, in crosses between a naked-seeded and a fuzzy-seeded upland cotton or between a nearly naked seeded and a fuzzy-seeded Egyptian cotton the first-generation plants were all naked-seeded or nearly so, and the second generation segregated approximately in a ratio of 3 naked (or nearly so) to 1 fuzzy (*19*, *29*, *30*). But in crosses between upland cottons having the seeds completely fuzzy and Egyptian cottons having the seeds only partly covered with fuzz, the F_1 plants all had the seeds completely covered and F_2 showed an unbroken range between the two extremes (*17*, *29*, *30*).

DESCRIPTION OF THE CHARACTER "SHORT BRANCH"

A striking variant was discovered by Byron Reddin of Tempe, Ariz., in his field of Pima Egyptian cotton in 1924. This plant was characterized by having all its fruiting branches reduced to only one developed internode. Seeds produced by naturally pollinated flowers on this individual were planted at the United States field station at Sacaton, Ariz., in 1925, and approximately half of the nine plants that came up resembled the original variant. The others had fruiting branches with more than one internode, although shorter than in typical Pima plants. Evidently many of the ovules on the mother plant of 1924 had been cross-fertilized by pollen from the surrounding normal plants.

Flowers were bagged on a plant in this progeny which resembled the original variant, and from the resulting self-fertilized seeds a progeny of five individuals was grown in 1926. All of these plants bore only reduced (1 internode) fruiting branches; and the inbred progenies successively descended from one of them, comprising 8 individuals in 1927, 11 in 1928, and 4 in 1929, expressed the character uniformly. It may be concluded that the original variant of 1924 and the plant selected for selfing in 1926 were homozygous for fruiting branches reduced to one developed internode.

The fact that the cotton plant bears two kinds of branches, vegetative branches and fruiting branches, was pointed out by Cook (*5*, *p. 13–31*). Cook showed that the vegetative branches arise from either axillary or extra-axillary buds on the main stem, usually near its base. The fruiting branches arise only from extra-axillary buds, one of which is produced normally at each node of the main stem (primary fruiting branches) and one at each node of the vegetative branches (secondary fruiting branches). The flowers ordinarily develop from extra-axillary buds on the fruiting branches, one at each node. There are, however, many deviations from this general plan of development, as Cook has shown.

None of the fruiting branches of the plants grown at the Sacaton station had more than one developed internode, at the summit of which were borne from one to three flowers. Where only one flower was borne, the appearance was as if a greatly elongated pedicel or flower stalk had sprung directly from a node of the main stem. Where

one or two additional flowers appeared, they may have developed from accessory buds of the same internode, or possibly each represented an additional internode, so greatly shortened as not to be discernible.[4] Occasionally the pedicels of two or three of these flowers were fused together (fasciated). Frequently two or three of these reduced fruiting branches arose from the same node of the main stem, the additional ones presumably by development from an axillary bud or from an axillary and an accessory bud, in addition to the single extra-axillary bud from which the fruiting branch at each node of the stalk normally develops.[5] The shortened fruiting branches were more nearly erect than the branches of normal Pima plants. This difference was due perhaps to the absence of the mechanical stimulus exerted by the heavier load of bolls on normal branches.

The occasional occurrence of a small bractlike leaf on the reduced branch, such as may be seen near the top of the right-hand plant in Figure 1, proves that the structure can not be regarded as a simple pedicel. Similar occurrences in Egyptian cotton plants were noted by Cook (*5, p. 24*), who wrote:

> Small bractlike leaves or stipules are occasionally present, even on straight stems, and sometimes the joint between the branch proper and the true stem or pedicel of the boll remains distinct, even when there are no leaves or stipules.

McLachlan (*25, p. 11*), referring to fruiting branches reduced to one or two internodes which sometimes appear to arise in an axillary position at the same node of the main stem with a normal, extra-axillary fruiting branch, states:

> At the lowest nodes where such branches appear they normally bear a leaf opposite the flower bud, but at higher nodes the leaf may be abortive and the branch appear as a very long flower stem borne directly on the main stalk of the plant.

The young plants illustrated in Figure 1 have produced only single flower buds at the summits of the short branches, but otherwise they show very well the striking character which distinguishes the variant from the normal Pima type. The latter is illustrated in Figure 2, representing a plant of corresponding age with fruiting branches extending to five developed internodes. It will be noted that the normal plant bears a leaf as well as a flower at each node of the fruiting branch (fig. 2), while the shortened fruiting branches of the variant are leafless or practically so. (Fig. 1.)

INHERITANCE OF SHORT BRANCH IN A CROSS WITH NORMAL BRANCH

In 1926 emasculated flowers on a plant of a Pima family having normally developed fruiting branches were pollinated with pollen from a plant in the first inbred progeny of the short-branch family. The normal Pima parent was of a progeny representing the twelfth successive inbred generation. For 12 years this family (PH–8) and others descended from plant P–1, self-pollinated in 1914, had furnished the parents representing typical Pima in crosses made for studying

[4] Referring to a condition sometimes found in the fruiting branches of the so-called "cluster cottons" (upland type), Cook states (*5, p. 18*): "The leaf buds that normally continue the growth of the branches are sometimes replaced by flower buds, or adjacent leaf buds may be aborted and fall off, so that the branch soon ends with a flower or a boll and no more joints can be added."

[5] A similar situation is described by Cook (*5, p. 16*) as follows: "Sometimes the normal extra-axillary fruiting branch is also replaced by a single flower bud, so that three flower buds may appear to come from each of the nodes of the main stem instead of the more normal complement of a limb and a fertile branch."

the inheritance of various characters (*17*, *18*). It had been under close observation during all this time and had shown no indication of the short-branch character. The same was true of the progenies

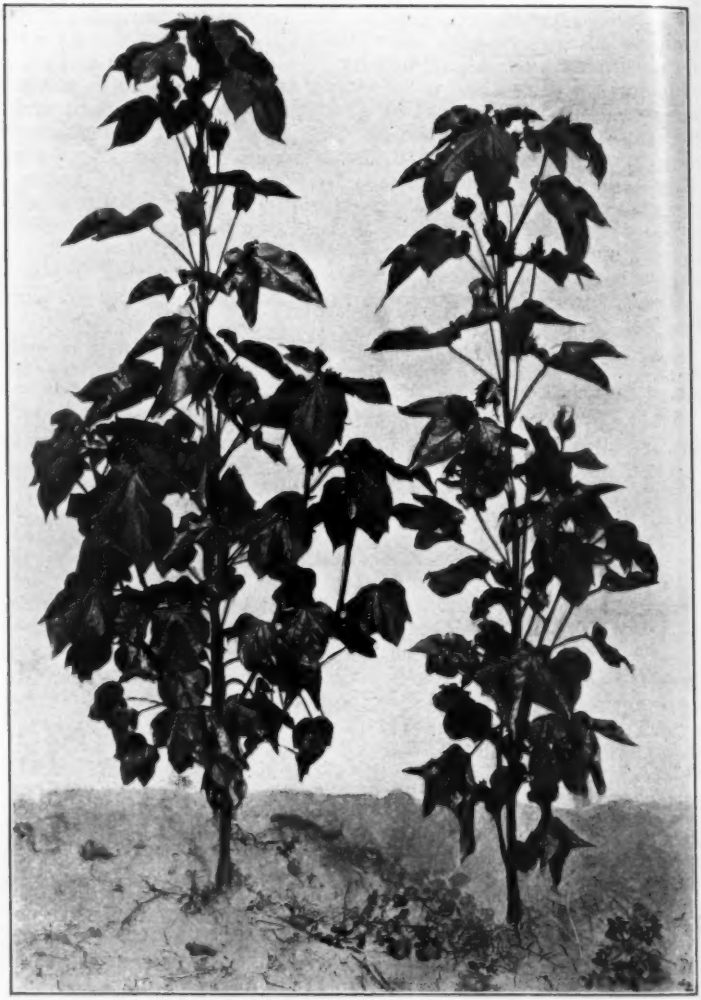

FIGURE 1.—Two individuals of the short-branch family of Pima cotton grown in 1928, representing the fourth generation of descent from the original selection. The fruiting branches are reduced to a single internode and are nearly erect. (Photographed July 31)

representing the thirteenth, fourteenth, and fifteenth inbred generations of family PH–8, grown in 1927, 1928, and 1929. In the 1928 progeny none of the 12 plants had fewer than five well-developed internodes on the longest fruiting branch. On the other hand,

the 11 individuals of the short-branch parental progeny grown in 1928 and the 4 individuals of the corresponding progeny grown in 1929 bore no fruiting branches having more than one internode.

An F_1 progeny of the cross was grown in 1927, but only two plants survived. Most of the fruiting branches on these plants were reduced

FIGURE 2.—A typical plant of Pima cotton (family PH-8) of the same age as the plants shown in Figure 1 grown in 1928. The longer fruiting branches have five well-developed internodes and are much less nearly erect than in the short-branch plants. (Photographed July 31)

to a single internode, but some of the branches on both plants had two well-developed internodes and on one of the plants a branch of three internodes was produced. The branches that had two or three internodes were more spreading, hence more nearly normal in direction, than the 1-internode branches.

An F_2 progeny of 48 individuals was grown in 1928 from seed produced by strictly self-pollinated (bagged) flowers on one of the F_1 plants. These plants were classified on the basis of the number of internodes of the fruiting branch which had the greatest number of internodes on the plant in question. The resulting frequency distribution is given in Table 1.

TABLE 1.—*Classification of the F_2 progeny of the cross "normal branch × short branch," 1928*

Number of plants	Frequency distribution for number of internodes of the fruiting branch having the highest number on the plant in question						Classification		
	1	2	3	4	5	6	Short (1 internode)	Intermediate (2 or 3 internodes)	Normal (4 to 6 internodes)
48	14	20	8	1	2	3	14	28	6

The classification in the last three columns of Table 1 was made on the assumptions that all individuals having no fruiting branches with more than 1 internode were homozygous for short branch, that all individuals on which the longest fruiting branch comprised not fewer than 4 internodes were homozygous for normal branch, and that the remaining individuals, which resembled the F_1 plants in having not more than 2 or 3 internodes on the longest fruiting branch, were heterozygous. On the assumption of a 1:2:1 ratio, the numbers expected in a population of 48 are 12:24:12, while the numbers observed were 14:28:6. χ^2 for the departure from the assumed ratio is 4.000, and taking $n\ (n-1)$ as 2, the value of P by Fisher's table of χ^2 (*6, Table 3*) is approximately 0.15. The indication, therefore, is that the short-branch character is conditioned by a single gene. The fact that the F_2 plants classed as intermediate resembled the two F_1 individuals grown in 1927 leads to the conclusion that there is no dominance in the heterozygous condition.

To test these assumptions, F_3 progenies were grown in 1929 from seed produced by flowers bagged on eight of the F_2 plants of 1928. Two of these represented the short-branch class, four the intermediate class, and two the normal-branch class. The classification of these F_3 progenies is given in Table 2.

With the exceptions noted in the footnote to Table 2, the diagnosis of the F_3 plants was easily made. It is believed that the classification given in the last three columns is accurate, since it was arrived at by considering the plant as a whole and laying not too much stress on the single criterion of number of internodes in the longest fruiting branch. In accepting this classification, it is seen that the two F_2 individuals selected as short branch and the two F_2 individuals selected as normal proved homozygous, there having been no segregation in their F_3 progenies. On the other hand, all four of the F_2 individuals which had been selected as intermediate were heterozygous, having produced F_3 progenies in which all three classes were represented.

The numbers of plants in the individual F_3 progenies are too small to give consequence to the ratios, taken singly. It is permissible, however, to combine them as one population, since they are all descended from a single cross and from a single self-fertilized F_1 plant.

On this basis, in a population of 56, there were 15 short-branch, 31 intermediate, and 10 normal-branch plants, whereas the numbers to have been expected with a 1:2:1 ratio are 14, 28, and 14. χ^2 of the departure from the expected ratio is only 1.537, and the indicated value of P is approximately 0.45. The chances therefore are about even that the departure is due to random sampling and would disappear if a larger population were available for classification.

TABLE 2.—*Classification of the eight F_2 progenies of the cross "normal branch × short branch," 1929*

F_2 progeny of F_1 No.	Number of plants	Classification of F_2 parent	Frequency distribution of F_2 for number of internodes of the fruiting branch having the highest number on the plant in question							Classification of F_2 on the same basis as F_2 *		
			1	2	3	4	5	6	7	Short	Intermediate	Normal
6	14	Short	14							14	0	0
14	15	do	14	1						15	0	0
5	14	Intermediate	7		3	2	1	1		7	5	2
8	14	do	4	1	1	4	3	1		4	6	4
11	15	do	3	4	4	2	1		1	3	11	1
15	13	do	1	1	6	2	2	1		1	9	3
21	14	Normal				4	6	4		0	0	14
35	14	do				1	8	5		0	0	14

* With the following exceptions: All individuals in the segregating F_2 progenies 5, 8, 11, and 15 which had 4 internodes on the longest fruiting branch also bore typical short branches of 1 internode only. The same was true of the individual in progeny 15 which had 7 internodes on the longest fruiting branch, the branch in question having been abnormal and partly metamorphosed into a vegetative branch. These 11 plants were classed as intermediate and only those individuals in the 4 segregating progenies which had 5 or 6 internodes on the longest fruiting branch were classed as normal. None of the latter plants bore any short (1-internode) branches, and thus they resembled all of the plants in the normal nonsegregating progenies 21 and 35. In progeny No. 15 one plant had several branches of 2 internodes. Since the other branches had only 1 internode, and since the 2-internode branches were otherwise of short-branch character, it seems reasonable to classify this individual as short and to regard the progeny in which it was the only aberrant plant as homozygous for short branch, notwithstanding the fact that no fruiting branches of more than 1 internode have been observed on any of the inbred descendants of the original short-branch plant.

The evidence from the third-generation populations strengthens materially the conclusion that short branch originated by mutation of a single gene. It may be added that no indication of linkage of this gene with any other has yet been discovered. So far as has been observed, the inbred descendants of the original short-branch plant and the segregates of this class in F_2 and F_3 of the cross are quite fertile and do not differ from the normal Pima family in any characters not attributable to the gene for short branch.

SUMMARY

Few characters of the cotton plant are known to be inherited in a definitely alternative manner. An addition to the short list of such characters, short branch, is described in this paper.

A plant showing the character was discovered in 1924 in a field of Pima Egyptian cotton in Arizona. The fruiting branches of this plant were reduced to a single internode which was leafless or practically so. Frequently two or three of these branches appeared at the same node of the main stem. From one to three flower buds were borne at the summit of the single internode. The character has been expressed uniformly in the inbred descendants of the original short-branch plant during four generations.

A cross was made in 1926 between a plant of an inbred Pima family having normal fruiting branches comprising usually five or more internodes and one of the short-branch plants. Only two plants survived in the F_1 progeny of the cross, and these showed an intermediate condition, bearing fruiting branches of from one to three internodes.

In the second generation there was segregation into three classes—short branch, intermediate, and normal or long branch. The proportions of the several classes suggested a 1:2:1 ratio, indicating absence of dominance in the heterozygous condition.

Third generation progenies were grown in 1929, representing two individuals of each of the presumably homozygous classes in F_2, and four individuals of the presumably heterozygous (intermediate) class. The short-branch and the normal F_2 individuals bred true, while all of the F_2 plants selected as being probably heterozygous produced segregating progenies. Taking the four segregating F_3 progenies as one population, the departure from a ratio of 1 short branch : 2 intermediate : 1 normal was insignificant.

The evidence seems complete that the expression of the character "short branch" depends upon a single gene and that there is no dominance in respect to this character.

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RELATION BETWEEN MOISTURE CONTENT OF THE WOOD AND BLUE STAIN IN LOBLOLLY PINE [1]

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INTRODUCTION

It is common knowledge that green sap lumber will become badly discolored by staining fungi unless the moisture content of the wood is reduced by drying the lumber at a fairly rapid rate. At certain seasons of the year, varying in different regions of the country, the general weather conditions are distinctly unfavorable for drying lumber. During such seasons blue staining is inevitable unless the lumber is treated with chemicals that prevent the development of the staining organisms, or unless drying is speeded up artificially by the use of dry kilns. At all seasons of the year control of the moisture content of the lumber is the most important phase of blue-stain prevention.

Neither the upper limiting moisture content at which the staining fungi will grow nor the optimum moisture content for their growth is of immediate interest from a practical standpoint in handling lumber. The lower limiting moisture content is, however, of paramount importance. Münch [3] determined that this lower limiting moisture content was approximately 28 per cent, based on the oven-dry weight of the wood, for *Pinus sylvestris* in Europe. The purpose of the present paper is to give the results of a series of accurately controlled tests to find the moisture content of the sapwood of one of the native American woods, loblolly pine (*P. taeda* L.), below which blue stain would not occur.

METHOD OF EXPERIMENTATION

The wood used for the tests came from a second-growth tree felled near Mize, Miss., in late November, 1925. A short log was cut from the bole at a point about 7 feet from the ground. The ends of the log were coated with heavy paint to retard moisture loss. The log was then shipped to Madison, Wis., where it was stored in the open for about four months until it was cut into test sticks. During this storage period little, if any, evaporation of moisture took place. In fact, the water in the wood froze and remained in a solid state until the log was cut in the mill. Except for a small central core of heartwood, the log was all sapwood. So far as could be determined, no infection with blue-stain organisms or wood-destroying fungi took place between the time of felling and the time of test.

The test sticks were prepared as follows: A bolt 2½ feet long was cut from the butt end of the log. The lower 6 inches of this bolt

[1] Received for publication May 2, 1930; issued September, 1930.

[2] In cooperation with the Forest Products Laboratory, Forest Service.

[3] MÜNCH, E. DIE BLAUFÄULE DES NADELHOLZES. Naturw. Ztschr. Forst u. Landw. 5: 531–73, illus., 1907; 6: 32–47, [297]–323, illus., 1908.

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were cut off and discarded. The freshly exposed lower end of the bolt was stained with a dye to facilitate the orientation of the individual boards as they came from the saw. The bolt was then quarter-sawed into nominal 1-inch boards. Every board was numbered as it came from the saw.

Each board was dressed in an accurate planer to a thickness of three-quarters of an inch. The sapwood of the boards was then laid off into test sticks, each of which was properly numbered. The test sticks were cut out carefully with a fine-tooth saw so that their width was as near three-fourths of an inch as possible. By this method a large number of sticks approximately 2 feet long and three-fourths by three-fourths inch in cross section were obtained.

These sticks were then cross piled openly in a conditioning room where the relative humidity was maintained at approximately 60 per cent. The relative humidity varied slightly, the lowest point reached being 58 and the highest 72 per cent. The temperature in the room remained close to 80° F., reaching a low point of 75° for only a short period. Preliminary tests showed that the wood dried so quickly under these conditions that after four days the moisture

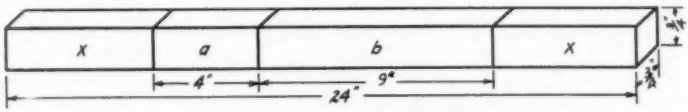


FIGURE 1.—Diagram showing the method used in cutting out the 4-inch moisture samples (*a*) and the 9-inch test sticks (*b*). The end pieces marked *x* were discarded

content had been reduced to approximately 14 per cent. This moisture content was known to be too low to support the growth of blue-stain fungi. Arrangements were therefore made to prepare some of the sticks for inoculation as soon as practicable after they were cut, and to remove others from the drying room for inoculation at short intervals after they had been placed therein. This procedure proved satisfactory. A series of test sticks with moisture content ranging from approximately 124 per cent to 14 per cent moisture content, oven-dry basis, was obtained.

The individual test sticks were cut from the 2-foot blanks, as shown in Figure 1. The middle part of each blank was laid off with the aid of a template into a 4-inch stick (fig. 1, *a*) to be used for determining moisture content and specific gravity, and a 9-inch test stick (fig. 1, *b*) to be used for inoculation. The 4-inch stick and the 9-inch stick were given the same identifying number and then cut. The pieces cut from the ends of the blank (fig. 1, *x*) were discarded.

The 4-inch and 9-inch sticks were placed in test tubes stoppered with cotton and then autoclaved 30 minutes at atmospheric pressure at a temperature of 100° C. After cooling, the 4-inch sticks were weighed. They were then dried to constant weight at 100° to 105° and weighed again. The percentage of moisture content, oven-dry basis, was later determined in the usual manner. Immediately after the dry weight was taken the dry samples were plunged into melted paraffin to prevent absorption of moisture. The volumes of the paraffin-coated sticks were determined by immersion in water in a finely graduated glass cylinder. The specific gravity of the sticks was

calculated on the basis of the oven-dry weight and the oven-dry volume.

It may be pointed out that a certain assumption is necessary in applying the results of the moisture content and specific gravity determinations made on the 4-inch sticks to the 9-inch test sticks. This assumption is that the moisture content of the smaller and larger pieces bearing like identifying numbers was the same after the samples had been autoclaved. The error in applying the specific gravity value of the smaller stick to the larger stick can be considered negligible. There may, however, have been a slightly larger loss in moisture content during autoclaving for the 4-inch stick than for the 9-inch test stick. The actual difference in the moisture content of any two like samples, however, is believed to have been slight, and it has been disregarded.

As soon as they had cooled, the 9-inch test sticks were inoculated with a strain of *Ceratostomella pilifera* which had been isolated in August, 1924, from the sapwood of loblolly pine. The infected wood came originally from North Carolina, but it is not known whether the log became infected there, during transit, or after it arrived at Madison.

The spores used for the inoculations were ascospores removed by a sterile needle from the tips of perithecia produced in a test tube on a malt-agar culture medium. The method of inoculation was as follows: A heavy suspension of spores was made in a small quantity of sterile water, and the spores were then transferred to the four sides of the test stick by means of a long platinum needle. An effort was made to distribute the spores from the top to the bottom of each side of each stick. No water other than the minute drops which clung to the platinum needle was added to any of the tubes either at the time of inoculation or later, so that it may be assumed that the inoculation method did not result in any significant increase in the moisture content of the inoculated sticks. The tubes containing the test sticks were capped with cotton pads held in place by pieces of cotton cloth tied down with string. They were placed in an upright position in a room with an average temperature of 78° F., and relative humidity of 82½ per cent.

Within 24 hours after inoculation the mycelium of the fungus could be observed along the track of the needle in the sticks having a moisture content high enough to support growth. Where the moisture content was distinctly favorable the development of the mycelium was rapid, and the sticks were completely enveloped in 21 days. The white mycelium soon became gray, and perithecia appeared in a few days. Where the moisture content was less favorable the mycelium was less luxuriant, the change to the typical gray color was retarded, and the formation of the perithecia was delayed or prevented. On sticks with relatively low moisture content the mycelium was concentrated toward the lower half of the stick, the upper part of the stick remaining clear.

EXPERIMENTAL DATA

Table 1 is made up of values for 48 apparently representative test sticks, calculated from the respective 4-inch moisture samples. Specific gravity, moisture content, and relative distribution of air,

wood substance, and water, expressed as percentage on a volumetric basis, are shown, together with the actual figures from which these values were determined. The values in columns 10 to 17 were obtained by the method described by Hartig [4] and used by Münch [5] in his description of the air, wood substance, and water relations in *Pinus sylvestris*. The figure 1.531 for the density of the actual wood substance in grams per cubic centimeter used in column 10, was specifically determined by Stamm [6] from two samples of wood from the *P. taeda* under test, representing, respectively, material of low and high density. It differs but slightly from the value of 1.55 found by Hartig and used by Münch. Hartig's value 0.55, representing the percentage of water that would be absorbed by the wood substance, was accepted as approximately correct, and it was used to calculate the relative amounts of imbibition water and of free water shown in columns 16 and 17. These calculations give a somewhat higher value for the fiber saturation point than has been determined by experiment. By using the numbers under the column headings to represent the values in the columns themselves, excepting the values 1.531 and 0.55, the method of calculating the values may be explained as follows:

$$7=3-4;\ 8=\frac{7}{4};\ 10=\frac{4}{1.531};\ 11=10+7;\ 12=6-(10+7);\ 13=\frac{12}{6};$$

$$14=\frac{10}{6};\ 15=\frac{7}{6};\ 16=10\times 0.55;\ 17=7-16.$$

The volume values in column 6 were calculated from data obtained from 52 additional 4-inch samples cut from blank sticks that were not used in any part of the preceding inoculation test. These sticks were prepared, however, at the same time and in the same machines as those used for the inoculation test. The 4-inch samples were laid out with the same template and cut with the same saw. Careful measurements and weighing showed them to be representative. Their volumes were determined by measuring the size of the samples with a steel scale graduated to 0.01 of an inch. The 4-inch samples had a range in moisture content from 102 per cent to 11.2 per cent. These values were plotted against the measured volumes and a curve drawn from which the average volume at any given moisture content could be read directly. The approximate average volume values were then inserted in column 6. It is obvious that this method is arbitrary. The volume in column 6 opposite laboratory number 35, for example, is not the actual volume of the 4-inch sample numbered 35, but the average volume of a 4-inch sample with an approximate moisture content of 25.4 per cent (column 8). The maximum error introduced by the use of an average instead of an actual volume value appears to be approximately 2 per cent at the wetter end of the series and 4 per cent at the drier end of the series. The difficulties in obtaining actual volume values for the sterilized test sticks at the beginning of the test are so great that the use of the arbitrary average value seems to be

[4] HARTIG, R. ÜBER DIE VERTHEILUNG DER ORGANISCHEN SUBSTANZ, DES WASSERS UND LUFTRAUMES IN BAUMEN, UND UBER DIE URSACHE DER WASSERBEWEGUNG IN TRANSPIRIRENDEN PFLANZEN. 112 p. illus. Berlin. 1882. (Untersuchungen aus dem Forst-botanischen Institut zu Muchen II.)

[5] MÜNCH, E. Op. cit.

[6] STAMM, A. J. DENSITY OF WOOD SUBSTANCE, ABSORPTION BY WOOD, AND PERMEABILITY OF WOOD. Jour. Phys. Chem. 33: [398]–414, illus. 1929.

justified. The error, whatever it may be, is carried over into columns 12, 13, 14, and 15 and into the graphic representation of the values in the last three of these columns. (Fig. 2.)

TABLE 1.—*Specific gravity, moisture content, and relative proportions of air, wood substance, and water in the test sticks at the beginning of the test*

Stick designation in Plates 1 and 2	Laboratory No.	Wet weight	Dry weight	Specific gravity, oven-dry basis	Volume at beginning of test	Water content [a]	Moisture content, oven-dry basis [b]	Moisture content, wet-wood basis [b]	Dry weight [c] / 1.531	$\left(\frac{\text{Dry weight}}{1.531}\right)+(\text{water content})$ [d]	[Vol. c. c. at beginning of test] $-\left[\left(\frac{\text{Dry weight}}{1.531}\right)+(\text{water content})\right]$ [e]	Air, volume basis	Wood substance, volume basis	Water, volume basis	Imbibition water	Free water
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
		Gm.	*Gm.*		*C. c.*	*C. c.*	*Per cent*	*Per cent*	*C. c.*	*C. c.*	*C. c.*	*Per cent*	*Per cent*	*Per cent*	*Gm.*	*Gm.*
A	1	39.14	17.50	0.522	37.0	21.64	123.7	55.3	11.43	33.07	3.93	10.6	30.9	58.5	6.28	15.35
	2	40.00	18.47	.535	37.0	21.53	116.6	53.8	12.06	33.59	3.41	9.2	32.6	58.2	6.65	14.90
B	3	40.42	18.91	.556	37.0	21.51	113.8	53.2	12.35	33.85	3.14	8.5	33.4	58.1	6.80	14.71
	4	38.11	17.85	.541	37.0	20.26	113.5	53.2	11.65	31.91	5.09	13.8	31.5	54.7	6.41	13.85
C	5	36.40	17.80	.524	37.0	18.60	104.5	51.1	11.62	30.22	6.78	18.3	31.4	50.3	6.39	12.21
	6	38.00	18.75	.551	37.0	19.25	102.7	50.6	12.25	31.50	5.50	14.9	33.1	52.0	6.74	12.51
D	7	37.51	18.92	.573	37.0	18.59	98.3	49.6	12.35	30.94	6.06	16.4	33.4	50.2	6.80	11.79
	8	38.40	19.60	.613	37.0	18.80	95.9	48.9	12.80	31.60	5.40	14.6	34.6	50.8	7.04	11.76
E	9	38.65	20.17	.630	37.0	18.48	91.6	47.8	13.16	31.64	5.36	14.5	35.6	49.9	7.24	11.24
	10	36.91	19.56	.630	37.0	17.35	88.7	47.0	12.76	30.11	6.89	18.6	34.5	46.9	7.02	10.33
F	11	36.53	19.36	.569	37.0	17.17	88.7	47.0	12.65	29.82	7.18	19.4	34.2	46.4	6.96	10.21
	12	37.10	19.81	.619	37.0	17.29	87.3	46.6	12.94	30.23	6.77	18.3	35.0	46.7	7.12	10.17
G	13	33.33	19.10	.538	37.0	14.38	74.5	42.7	12.47	26.70	10.30	27.8	33.7	38.5	6.86	7.37
	14	33.70	19.50	.565	37.0	14.20	72.8	42.1	12.74	26.94	10.06	27.2	34.4	38.4	7.01	7.19
	15	32.43	19.20	.565	37.0	13.23	68.9	40.8	12.54	25.77	11.23	30.3	33.9	35.8	6.90	6.33
	16	30.02	18.04	.547	37.0	11.98	66.4	39.9	11.77	23.75	13.25	35.8	31.8	32.4	6.47	5.51
H	17	32.22	19.42	.598	37.0	12.80	65.9	39.7	12.69	25.49	11.51	31.1	34.3	34.6	6.98	5.82
	18	31.38	19.90	.603	37.0	11.48	57.7	36.6	13.00	24.48	12.52	33.9	35.1	31.0	7.15	4.33
	19	31.60	20.26	.588	37.0	11.32	55.8	35.8	13.24	24.55	12.44	33.6	35.8	30.6	7.28	4.06
	20	27.63	18.41	.558	37.0	9.22	50.1	33.4	12.03	21.25	15.75	42.6	32.5	24.9	6.62	2.60
	21	29.53	19.68	.596	37.0	9.85	50.1	33.3	12.85	22.70	14.30	38.7	34.7	26.6	7.06	2.79
I	22	22.80	15.62	.504	37.0	7.18	46.0	31.5	10.20	17.38	19.62	53.0	27.6	19.4	5.61	1.57
	23	24.21	16.59	.510	37.0	7.62	45.9	31.5	10.83	18.45	18.55	50.1	29.3	29.6	5.96	1.66
J	24	27.00	18.57	.563	37.0	8.43	45.4	31.2	12.14	20.57	16.43	44.4	32.8	22.8	6.68	1.75
	25	27.80	19.37	.587	37.0	8.43	43.5	30.3	12.65	21.08	15.92	43.0	34.2	22.8	6.96	1.47
	26	26.92	19.28	.567	37.0	7.64	39.6	28.4	12.60	20.24	16.76	45.3	34.0	20.7	6.93	.71
K	27	24.72	18.24	.570	37.0	6.48	35.5	26.2	11.90	18.38	18.62	50.3	32.2	17.5	6.54	−.06
	28	22.10	16.90	.504	36.9	5.20	30.8	23.5	11.04	16.24	20.66	56.0	29.9	14.1	6.01	−.87
L	29	23.03	17.93	.552	36.9	5.10	28.4	22.1	11.70	16.80	20.10	54.5	31.7	13.8	6.44	−1.34
	30	22.00	17.17	.536	36.8	4.83	28.1	22.0	11.21	16.04	20.76	56.4	30.5	13.1	6.16	−1.33
	31	23.35	18.36	.574	36.8	4.99	27.2	21.4	12.00	16.99	19.81	53.8	32.6	13.6	6.60	−1.61
M	32	23.91	18.95	.557	36.8	4.96	26.2	20.7	12.38	17.34	19.46	52.9	33.6	13.5	6.81	−1.85
	33	22.92	18.21	.560	36.7	4.71	25.9	20.5	11.90	16.61	20.09	54.8	32.4	12.8	6.54	−1.83
	34	21.67	17.24	.530	36.7	4.43	25.7	20.4	11.25	15.68	21.02	57.2	30.7	12.1	6.19	−1.76
N	35	22.65	18.07	.548	36.7	4.58	25.4	20.2	11.80	16.38	20.32	55.4	32.1	12.5	6.49	−1.91
	36	23.67	18.90	.591	36.7	4.77	25.2	20.2	12.35	17.12	19.58	53.4	33.6	13.0	6.80	−2.03
O	37	23.00	18.44	.576	36.6	4.56	24.7	19.8	12.05	16.61	19.99	54.6	32.9	12.5	6.62	−2.06
	38	24.23	19.52	.622	36.6	4.71	24.1	19.4	12.75	17.46	19.14	52.3	34.8	12.9	7.01	−2.30
	39	24.04	19.39	.570	36.5	4.65	24.0	19.3	12.65	17.30	19.2	52.6	34.7	12.7	6.96	−2.31
P	40	23.17	18.69	.623	36.5	4.48	24.0	19.3	12.20	16.58	19.82	54.3	33.4	12.3	6.71	−2.23
	41	24.28	19.75	.598	36.3	4.53	22.9	18.7	12.90	17.43	18.87	52.0	35.5	12.5	7.10	−2.57
	42	19.88	16.21	.514	36.3	3.67	22.6	18.5	10.59	14.26	22.04	60.7	29.2	10.1	5.82	−2.15
Q	43	23.06	18.91	.610	36.3	4.15	22.0	18.0	12.35	16.50	19.80	54.6	34.0	11.4	6.80	−2.65
R	44	19.95	16.49	.492	36.1	3.46	21.0	17.3	10.76	14.22	21.88	60.6	29.8	9.6	5.92	−2.46
	45	20.90	17.31	.525	36.0	3.59	20.7	17.2	11.30	14.89	21.11	58.6	31.4	10.0	6.22	−2.63
S	46	21.81	18.58	.562	35.4	3.23	17.4	14.8	12.14	15.37	20.03	56.6	34.3	9.1	6.68	−3.45
T	47	22.46	19.55	.611	34.9	2.91	14.9	12.97	12.77	15.68	19.22	55.1	36.6	8.3	7.02	−4.11
	48	22.30	19.56	.631	34.8	2.74	14.0	12.3	12.78	15.52	19.28	55.4	36.7	7.9	7.03	−4.29

[a] Equivalent to volume of water in cubic centimeters (temperature change disregarded).
[b] Computed from adjoining 4-inch segments.
[c] Equivalent to volume of wood substance in cubic centimeters.
[d] Equivalent to volume of wood substance plus volume of water in cubic centimeters.
[e] Equivalent to volume of air in cubic centimeters.

The points plotted on Figure 2 represent the values on a volumetric basis for the percentages of air, wood substance, and water that are given in Table 1. The curves were drawn by inspection. Two sets of moisture-content percentage values based on weight are listed on the x axis, and the percentage of moisture content based on volume is indicated on the y axis. The method of plotting used makes the relation among the three ways of stating moisture content fairly clear. However, since there may be many readers who are more or less

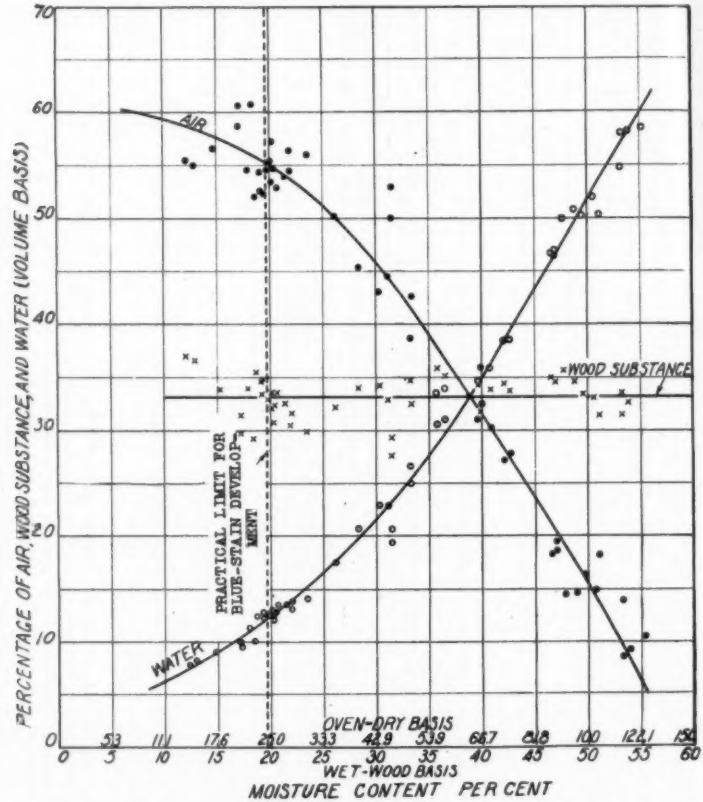


FIGURE 2.—Relative volumetric distribution of air, wood substance, and water in the test sticks at the beginning of the test

unfamiliar with the two more common methods of expressing moisture content on a weight basis, it has been thought best to include Figure 3. The curves in this figure show at a glance the relation between moisture-content percentages based on the weight of the oven-dry wood and moisture-content percentages based on the weight of the wet wood.

Table 2 gives a general description of the test sticks at the end of the test. The laboratory number and moisture content data are the

same as in Table 1. Tables 1 and 2, Figure 2, and Plates 1 and 2 should be studied together. Moisture content interpolations can be made with the help of Figure 3.

TABLE 2.—*Results of the experiments on the inoculation of loblolly pine of different percentages of moisture content with a strain of Ceratostomella pilifera*

Stick designation in Plates 1 and 2	Laboratory No.	Moisture content, oven-dry basis, at beginning of test [a]	Duration of test [b]	Appearance at end of test	
				Surface	Interior
		Per cent	*Days*		
A	1	123.7	36	Covered with mycelium, abundant perithecia.	Completely stained, dark.
	2	116.6	36	....do....	Do.
B	3	113.8	36	....do....	Do.
	4	113.5	21	....do....	Do.
C	5	104.5	35	....do....	Do.
	6	102.7	36	....do....	Do.
D	7	98.3	35	....do....	Do.
	8	95.9	35	....do....	Do.
E	9	91.6	36	....do....	Do.
F	10	88.7	35	....do....	Do.
	11	88.7	35	....do....	Do.
	12	87.3	35	....do....	Do.
G	13	74.5	34	....do....	Do.
	14	72.8	34	....do....	Do.
	15	68.9	34	....do....	Do.
	16	66.4	33	....do....	Do.
H	17	65.9	34	....do....	Do.
	18	57.7	34	....do....	Do.
	19	55.8	30	....do....	Do.
	20	50.1	33	....do....	Do.
I	21	50.1	33	....do....	Do.
	22	46.0	22	....do....	Do.
	23	45.9	22	....do....	Do.
J	24	45.4	33	Covered with white mycelium..	Do.
	25	43.5	31	....do....	Do.
K	26	39.6	33	Covered with light-gray mycelium.	Do.
	27	35.5	22	....do....	Do.
L	28	30.8	22	Lower 8¼ inches covered with mycelium.	Bottom 3 inches completely stained, medium.
	29	28.4	22	Lower 8 inches covered with mycelium.	Bottom 6½ inches completely stained, medium.
M	30	28.1	22	Lower 7 inches covered with mycelium.	Bottom 7 inches completely stained, medium.
	31	27.2	22	Lower 7½ inches covered with mycelium.	Bottom 2½ inches completely stained, medium.
	32	26.2	22	Lower 6½ inches covered with mycelium.	Bottom 1½ inches completely stained, medium.
	33	25.9	23	Lower 2 inches covered with mycelium.	Bottom 2 inches completely stained, medium.
	34	25.7	22	Lower 2½ inches covered with mycelium.	Bottom 2 inches completely stained, light.
N	35	25.4	22	Lower 4 inches covered with mycelium.	Bottom ½ inch completely stained, light.
	36	25.2	22	Lower 3 inches covered with mycelium.	Bottom ¾ inch completely stained, light.
O	37	24.7	22	Lower 1¾ inches covered with mycelium.	Bottom 2 inches completely stained, light.
P	38	24.1	22	Lower 1 inch covered with mycelium.	Bottom 1 inch completely stained, light.
	39	24.0	23	No mycelium evident.	Unstained.
	40	24.0	22	White and gray mycelium on bottom ½ inch.	Do.
	41	22.9	22	White and gray mycelium on bottom 1 inch.	Do.
	42	22.6	21	....do....	Do.
Q	43	22.0	22	....do....	Bottom ½ inch completely stained, light.
R	44	21.0	21	White mycelium on bottom ½ inch.	Unstained.
	45	20.7	21	....do....	Do.
S	46	17.4	32	Scant mycelium on bottom 2½ inches.	Do.
T	47	14.9	32	Scant mycelium on bottom ½ inch.	Do.
	48	14.0	32	....do....	Do.

[a] Computed from adjoining 4-inch segments.

[b] Two experiments were run; the first closed after 30 to 36 days and the second after 21 to 22 days, a period found sufficient for complete interior staining.

The distribution of moisture in nine representative test sticks at the end of the test is shown in Table 3.

TABLE 3.—*Distribution of moisture (per cent) in some of the test sticks at the end of the tests*

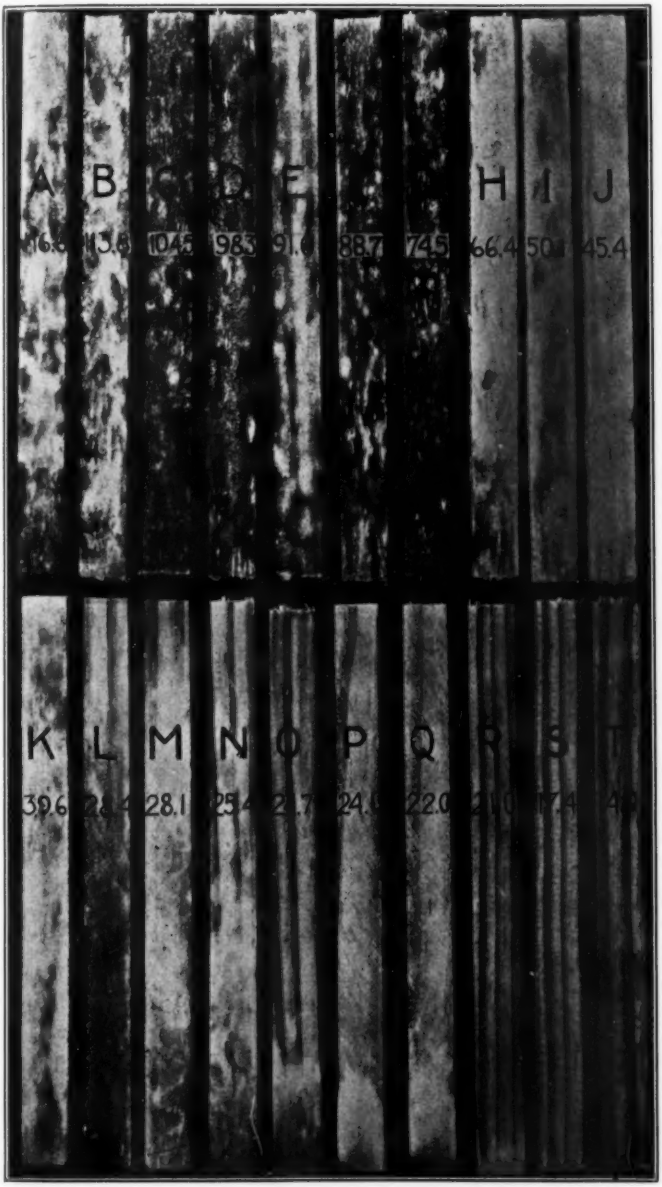
Laboratory No.	Moisture content of 1-inch segments of test sticks, oven-dry basis									Moisture content of whole stick, oven-dry basis.			Duration of test (days)
	Bottom segment 1	Segment 2	Segment 3	Segment 4	Segment 5	Segment 6	Segment 7	Segment 8	Top segment 9	At beginning of test[a]	At end of test[b]	Reduction during test[c]	
9453	23.9	23.1	22.4	20.0	20.8	19.7	18.4	15.0	14.8	22.6	19.8	2.8	21
9416	37.6	41.1	39.3	39.0	38.6	39.2	40.0	38.5	40.1	45.9	39.3	6.6	22
9415	32.3	35.4	35.5	33.5	34.5	34.0	32.6	30.6	27.4	35.5	32.8	2.7	22
8437	40.6	42.8	44.5	42.4	39.7	42.6	40.2	42.0	39.6	55.8	41.6	14.2	30
8433	39.0	40.0	37.8	36.4	37.5	36.2	34.5	35.0	30.6	50.1	36.3	13.8	39
9475	29.0	28.2	26.6	25.4	22.9	22.3	20.3	17.6	16.0	25.7	23.1	2.6	22
9414	34.4	32.1	31.1	27.5	27.8	26.8	23.7	20.6	17.4	27.2	26.7	.5	22
9423	32.7	33.5	31.6	30.6	29.1	26.8	24.5	21.2	17.1	30.8	27.5	3.3	22
9413	45.8	43.9	42.8	41.0	43.0	42.7	41.6	42.3	39.1	46.0	42.5	3.5	22

[a] Assumed to be same as that found in an adjacent 4-inch segment from the same stick.
[b] Determined from total dry weight and total water from the 9 segments.
[c] Difference between the two preceding figures.

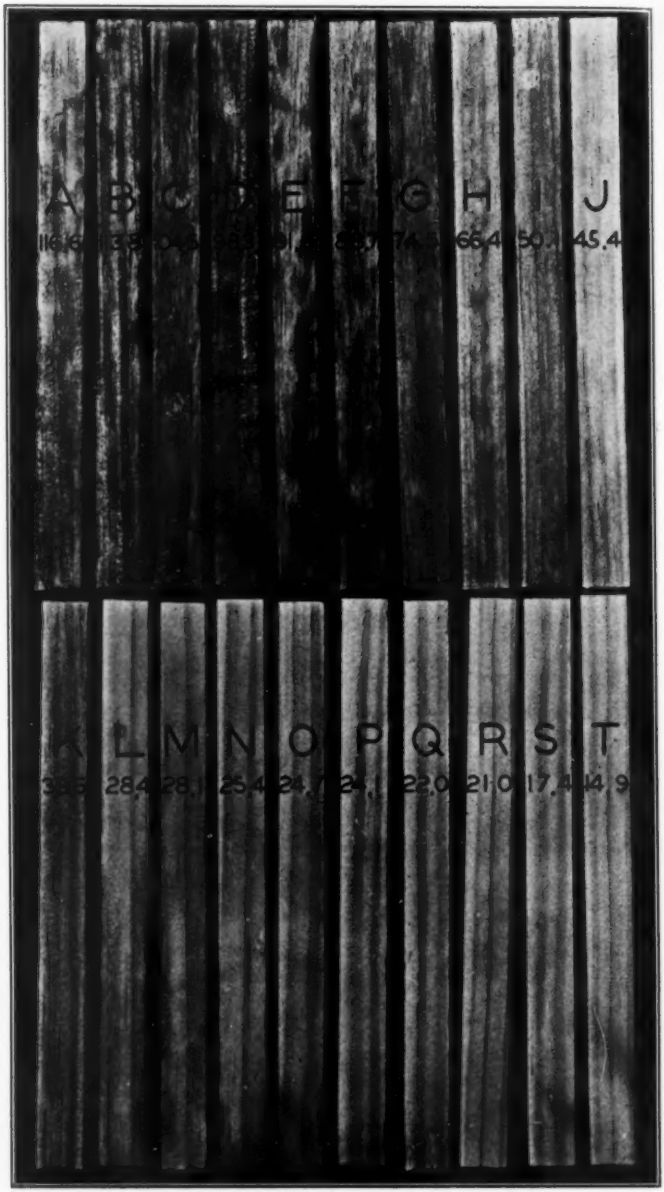
DISCUSSION OF RESULTS

Sticks A to G in Plate 1 are covered with gray mycelium. Sticks H to K are covered with mycelium that is not so dark. Stick L shows approximately 1 inch of clear wood at the top, and from stick M to stick R there is a rapid falling off in the height reached by the mycelium. On stick R the mycelium is reduced to little more than a trace at the very bottom. The moisture content values of sticks O to R at the beginning of the test ranged from 24.7 per cent down to 21.0 per cent, oven-dry basis. Stick S showed a scant white mycelium at its base which disappeared when the stick was handled. Stick T showed a white mycelium at its base, so tenuous and sparse that it had disappeared before the end of the experiment. (Pl. 1.) Data on the distribution of air, water, and wood substance in these sticks are given in Table 1.

After the negative for Plate 1 was taken the sticks were sawed lengthwise. The freshly exposed surface of one-half of each stick was then planed, and the planed sticks were arranged (pl. 2) in the same order as they appear in Plate 1. This method allows easy comparison between the exterior and the interior appearance of the sticks. It is evident from Plate 2 that sticks A to G were heavily stained throughout. Beginning with stick H there is a steady diminution in the amount of stain, with the last visible trace of discoloration showing at the bottom of stick Q. On the evidence presented in these two plates it might be concluded that the lower limiting moisture content, below which the staining organism would not develop, was approximately 21 to 22 per cent, oven-dry basis. Table 2 shows that development of mycelium was very evidently retarded on sticks which had a moisture content at the beginning of the test of 24 per cent or less. The data in Table 3 help to explain these results.



Twenty representative test sticks at the end of the test, arranged in series to show the relation between moisture content and the development of mycelium on the surfaces of the sticks. The moisture-content values of the sticks at the beginning of the test are marked on them. Complete data are shown in Tables 1 and 2



The same 20 sticks that appear in Plate 1, split to show the stain inside of the test sticks

Stick 42, for example, had a moisture content of 22.6 per cent at the beginning of the test. At the end of the test the moisture content was found to be 19.5 per cent; that is, there was a loss of 3.1 per cent during the 21 days the test was run. Examination of adjoining 1-inch segments of the stick showed that there was a definite moisture gradient in the stick with a high moisture content of 23.9 per cent in the bottom inch and a low moisture content of 14.8 per cent in the top inch. At just what point in the course of the test this gradient was established is not known. It can be safely assumed that the gradient was not constant. How the gradient was established seems reasonably clear. The loss in moisture content shown in Table 3 must have been

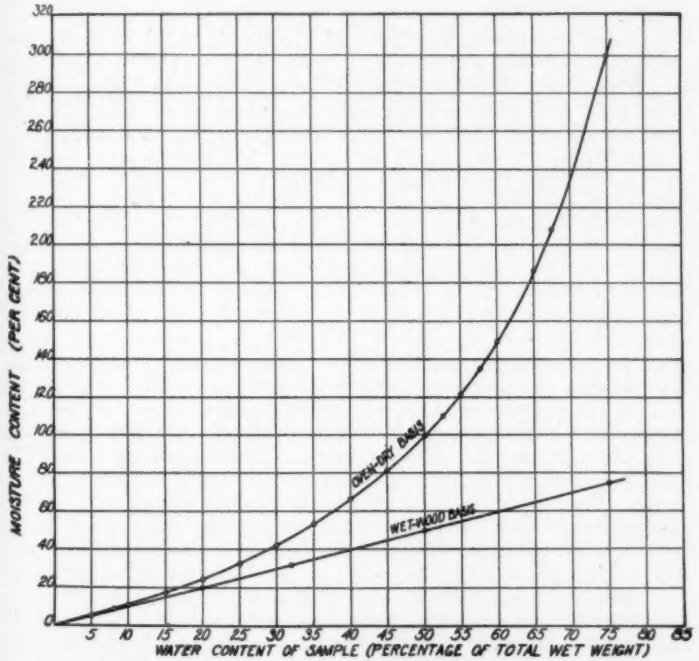


FIGURE 3.—Relation between moisture content percentages based on the weight of the oven-dry wood, and moisture-content percentages based on the weight of the wet wood

caused by evaporation through the cotton pad and cloth. One would therefore expect the upper end of the stick to be somewhat drier than the lower end. There is another reason, however, for the higher moisture content of the lower end. Very slight changes in temperature caused a condensation of moisture on the inside of the tubes. The moisture collected in drops and ran to the bottom of the tube. Daily observation showed that this moisture was absorbed by the wood at the lower end of the stick. The two processes, evaporation from the top of the stick and absorption at the bottom of the stick, resulted in the establishment of the moisture gradient. In stick 42 the moisture content near the bottom at the beginning of the test or soon there-

after may have been several points in excess of the 23.9 per cent found at the end of the test. The moisture content was evidently high enough to support the growth of the mycelium for a time, but not high enough to result in staining the interior of the stick. (Table 2.)

Table 3 shows that the gradient was sometimes steeper than in stick 42. In stick 34, for example, each of the lower 4 inches of the stick had a moisture content in excess of 25 per cent at the end of the test, although the moisture content for the whole stick was only 22.7 per cent. Table 2 shows that the lower 2½ inches of this stick were covered with mycelium and that the lower 2 inches of the stick were stained inside. At the beginning of the test this stick had a moisture content of 25.7 per cent. It may be compared with stick N in Plates 1 and 2.

In sticks similar to stick 20 or 23 (Table 3), which are comparable to sticks I and J in Plates 1 and 2, and in sticks with a still higher moisture content the mycelium acts as a sort of wet blanket covering the surface, and in such cases moisture gradients are not likely to be established in tests running for so short a period as the duration of these tests.

CONCLUSIONS

From the tests it appears that the sapwood of *Pinus taeda* at a moisture content of 24 per cent or less, oven-dry basis, will not support sufficient growth of this strain of *Ceratostomella pilifera* to produce staining of the wood. At a moisture content of 24 per cent, oven-dry basis, which is equivalent to 19.3 per cent on the wet-wood basis, approximately 53.5 per cent of the volume of the wood is air, 35 per cent is wood substance, and 12.5 per cent is water, assuming reasonable accuracy in the methods used for determining the values shown in Table 1. The dotted vertical line at 24 (oven-dry scale) in Figure 2 marks the limiting conditions described above. A moisture content of 24 per cent should not be considered as the absolutely fixed limit below which blue stain will not occur. It may be assumed, however, that the lower limiting value is somewhere near 24 per cent.

Münch [7] concluded that when air made up 61 per cent of the volume and when the wood substance lacked 7 per cent (oven-dry weight basis) of being saturated the development of *Ceratostomella coerulea* was practically inhibited in *Pinus sylvestris*. The moisture content, oven-dry basis, calculated for these conditions from the values given in his Table 1 was 28. He says, however, that the fungus developed in the lower third of his test block and that the upper part of the block remained unstained. There is at least a possibility, judging from the results of the tests on *P. taeda*, that the lower part of the block used by Münch also had a moisture content in excess of 28 per cent. At any rate he mentions the fact that evaporation from the lower part of the block was somewhat retarded. Considering the possible effect of the difference in species of wood and the strain of fungus used, the results obtained on *P. taeda* check very well with the results on *P. sylvestris*.

From the growth reactions of *Ceratostomella pilifera* in culture on *Pinus taeda* in the laboratory, and from field observations in the southern pine area, it appears very probable that blue-stain fungi are

[7] MÜNCH, E. Op. cit.

not likely to grow if the moisture content of the outside layers of cells is 24 per cent or below, provided the relative humidity is low. The moisture content of the interior wood might conceivably be higher than 24 per cent for a while at least; but if the moisture content of the outside of the wood is too low to support growth, a reduction in the amount of blue-stain infection is certainly possible. In excessively damp or rainy weather the outside layers of cells might have a moisture content of 25 per cent or more although the interior of the wood remained for a time somewhat drier. Under such conditions it is conceivable that blue-stain fungi might start growing. Unless the rainy weather were prolonged, as it is, of course, during the regular rainy season, the outside layers of cells would soon dry to below 24 per cent and the growth of the organisms would be stopped.

Attention is particularly drawn to the fact that the results presented in this paper were obtained on sapwood that had been submitted to a temperature of 100° C. for 30 minutes. Such steamed wood may be slightly different from sapwood that has not been so treated, due to the volatilization of some fractions of the resinous materials in the wood. No tests were made to determine this point. To avoid serious mold contaminations one is practically compelled to use wood that is disinfected at least superficially. The attempt was made in these tests to reduce the heat treatment to the shortest practicable period. The inoculation results are believed to approach those that would have been obtained on unsterilized *Pinus taeda* wood with an approximately equivalent moisture content, but no supporting data can be presented.

Field observations indicate some difference in the staining reactions of the different southern pines. For example, in certain sections it is commonly held that shortleaf pine stains more quickly and more severely than longleaf pine. If this be generally true, there appears to be considerable justification in applying the results of the tests on *Pinus taeda* in control experiments on the southern pines. Additional laboratory tests on these other pines with wood cut at various seasons of the year are necessary before it can be said more definitely what is the low moisture content that will inhibit the growth of blue-stain fungi in each species of wood. It seems wisest, therefore, to set a figure of 20 per cent moisture content as the practicable working limit and to consider the difference between this 20 per cent and the 24 per cent deduced from the tests as a reasonable margin or factor of safety.

THE ADSORPTION OF THE ANIONS OF ACID DYES BY SOIL COLLOIDS[1]

By J. G. SMITH, *Assistant Chemist*, and P. L. GILE, *Senior Chemist, Bureau of Chemistry and Soils, United States Department of Agriculture*

INTRODUCTION

Previous studies (*2*)[2] have shown that the capacity of soil colloids for adsorbing or exchanging inorganic cations varies roughly with the molecular silica-sesquioxide ratio of the colloid. This parallelism has been explained on the ground that the exchangeable bases are held in some sort of combination with acidic, silica, or alumino-silicate radicals and that the reactive surface of the colloid occupied by these radicals varies roughly with the gross content of SiO_2 (*1, p. 579*). It would seem to follow that those colloids having low silica-sesquioxide ratios and low exchangeable base capacities should have more or less aluminum or iron in the surface and that these colloids should therefore have appreciable adsorptive capacities for anions.

This idea that part of the surface of a soil colloid should be capable of combining with cations and part capable of combining with anions is not wholly supported by the available data. Although different soil colloids have appreciable and varying capacities for adsorbing cations from both neutral salt and basic dye solutions, they seem to have little if any capacity for adsorbing simple anions, such as Cl, NO_3, and SO_4, at least from neutral salt solutions. On the other hand, adsorptions of the complex anions of acid-dye solutions have been frequently reported.

In case anions of acid dyes are adsorbed, it would seem that the comparative capacities of a soil colloid for adsorbing acid and basic dyes should furnish a better characterization of the colloid than the single determination of the adsorption of a basic dye. It is therefore important to determine whether there is an acid dye that is appreciably adsorbed by soil colloids, quite aside from any evidence that may be afforded regarding the nature of the colloid surface. An investigation of the adsorptions of several acid dyes is described in this paper.

REVIEW OF LITERATURE

The idea that the adsorptive capacities of soil and other inorganic colloids for different dyes would throw light on the constitution of the materials is far from new. Sjollema (*15*) suggested in 1905 that the extent to which soil colloids adsorb methyl violet, naphthol yellow, Congo red, and alizarin would indicate the presence of silicic acid, hydrous aluminum oxide, and aluminum silicate, since artificial gels of these compounds adsorb these dyes in different degrees. However, he gave no data on the adsorption of soils. Later Hundeshagen (*11*) showed that the adsorption of different dyes helped to identify

[1] Received for publication June 6, 1930; issued September, 1930.
[2] Reference is made by number (italic) to Literature Cited, p. 413.

certain minerals. The use of dyes for indicating the nature of the adsorbent is also suggested by the work of Michaelis and Rona (*13*), Bechhold (*6*), Testoni (*16*), and Birutowitsch (*7*) with artificial gels. In these investigations it was found that silica gels adsorbed only basic dyes, whereas alumina and iron gels adsorbed only acid dyes. Recently, Croucher (*8*) showed that adsorption of the acid dye, Biebrich scarlet, by an artificial hydrous alumina gel increased with increasing acidity, while adsorption of the basic dye, iodine green, decreased with increasing hydrogen-ion concentration, the minimum adsorption of the combined dyes occurring at a pH of 6.5, the isoelectric point of the gel. Since soils high in alumina and iron showed a similar behavior, he suggested that a marked change in the proportions in which these dyes are adsorbed above and below pH 6.5 constitutes a test for the presence of hydrous alumina in soils.

Observations on the adsorption of acid dyes by soil materials have been made by several investigators, although most quantitative studies of dye adsorption by soils (*10*) and clays (*3*, *4*) have been conducted with the two basic dyes methylene blue and malachite green, which are highly adsorbed. Way (*17*) states that clays remove the coloring matter from cochineal solutions. According to Rohland (*14*), adsorption of the acid dye aniline blue by soils is comparable to that of the basic dyes crystal violet and Victoria blue; adsorption of the acid dyes orange and metanil yellow is also appreciable but much less than that of aniline blue. Bechhold (*6*) found that a clay and a permutite both adsorbed methylene blue strongly but took up none of the acid dye trypan blue and very little of the acid dye toluene red. Data on the adsorption of diamine sky-blue are given by Beaumont (*5*) for four soils. The quantities of this acid dye adsorbed were appreciable, but much less than the quantities of the basic dye methylene blue. Michaelis and Rona (*13*) report that a kaolin which adsorbed several basic dyes strongly did not adsorb any of the acid dyes potassium eosinate and ammonium picrate, but did adsorb some of the more colloidal dyes, Congo red and diamine fast red. Wilkinson and Hoff (*20*) give the adsorptions of six soils for two basic dyes and one acid dye, diamine blue 3 B, with and without the addition of acid. All dyes were appreciably adsorbed in the absence of acid, but the adsorption of diamine blue 3 B was especially increased by acid. Birutowitsch's data show that a "Floridin" soil adsorbed the acid dyes night blue, eosin, and Congo red, but not azo blue. Croucher (*8*) reports a marked adsorption of Biebrich scarlet (an acid dye) by soils high in iron and aluminum from solutions below pH 6.5. These results on the adsorption of acid dyes may be summarized as follows: Certain investigators found trypan blue, toluene red, potassium eosinate, and ammonium picrate not adsorbed by soils or similar materials; other investigators report adsorptions of cochineal, aniline blue, orange, metanil yellow, diamine sky blue, Congo red, diamine fast red, diamine blue 3-B, night blue, eosin, and Biebrich scarlet. The adsorption of some of these dyes was markedly increased by acidifying the solution.

Certain facts established regarding the adsorption of dyes by charcoal and by silicon, iron, and aluminum oxide gels have a bearing on the adsorption of dyes by soils. It was shown by Michaelis and Rona (*13*) that the capacities of the so-called "pure" inorganic gels for dyes

and simpler electrolytes are dependent on impurities of Cl, Na, etc., remaining in the adsorbent after purification. The adsorption consists of an exchange between the adsorbed ion and the ion of similar charge present as impurity in the adsorbent. Charcoal seems to be an exception to many adsorbents of electrolytes in that only part of its adsorption can be attributed to this exchange reaction. This conclusion that the adsorption of dye electrolytes by inorganic gels is an exchange reaction is substantiated by the work of Wood and Wooller (*21*) on several alumina gels, by the work of Weiser and Porter (*19, p. 1830*) on a chromic oxide gel, and by the work of Weiser (*18*) on alumina gels.

This generalization of Michaelis and Rona, that the adsorption of dyes is an exchange reaction, applies also to the adsorption by soils, of at least the basic dyes. Some years before the work of Michaelis and Rona, Ashley (*3, p. 58*) suggested that the adsorption of malachite green by clays was an exchange reaction "with the bases that stabilize the gel." Rohland (*14, p. 381*) also suggested that the adsorption of dyes by soils was often connected with exchange reactions. The data of Michaelis and Rona showed that kaolin takes up basic dyes in this way. And recently Anderson and Mattson (*2*) showed quantitatively that the adsorption of methylene blue by two widely different soils was an exchange reaction, at least up to a certain point. This was substantiated qualitatively by Wilkinson and Hoff (*20*) for several soils. The adsorption of basic dyes by soils is thus similar to the more thoroughly studied base exchange reaction with neutral salts.

If the facts established for the adsorption by soils are compared with the facts established for adsorption by artificial inorganic gels, an indication is obtained of how much acid dye soils should adsorb. Apparently the artificial gels adsorb basic dyes by exchange with cations and acid dyes by exchange with anions. Soils adsorb basic dyes by exchange with cations, and they would presumably adsorb the anions of acid dyes by exchange. Since soils seem to adsorb dye cations and the cations of simple salts to about the same extent, they would be expected to adsorb dye anions and simpler anions in equivalent amounts. Previous studies show, however, that soils adsorb only small quantities of the simple inorganic anions, if any; hence little if any adsorption of acid-dye anions would be expected. This conclusion is in harmony with the findings of certain investigators cited but is not in agreement with the data of others.

EXPERIMENTAL PROCEDURE

In the adsorption determinations, the soil was shaken with a dye solution and the unadsorbed dye was determined colorimetrically after the suspension had been clarified by centrifuging. All adsorption determinations were conducted with a 0.1 per cent dye solution which was made from a clear, 0.2 per cent stock solution. The stock solution was prepared by passing a dye solution through a Pasteur-Chamberland filter and adjusting to 0.2 per cent strength after determining the solid present in an aliquot evaporated on the steam bath. The weights of soil and volumes of dye used in different determinations were varied in such a way that about half of the dye supplied remained unadsorbed in each case; thus the adsorbed dye was in equilibrium

with an approximately 0.05 per cent solution. It was usually necessary to make several preliminary tests in order to determine the proper quantities of dye and soil to use. The soil and dye solution was shaken for 17 hours in an end-over-end shaker, and the dye remaining unadsorbed was then determined colorimetrically as follows: After the soil had settled somewhat, approximately 40 c. c. of the supernatant solution was withdrawn and centrifuged for 30 minutes, or until clear. In some cases a drop or two of dilute hydrochloric acid was added to facilitate the flocculation of clay. The color of the clarified solution was then read in a Dubosq colorimeter against that of a 0.05 per cent solution of the dye, similarly acidified when acid was used in clarification.

The soils used in this study were selected from a number of those utilized in different investigations of this laboratory. The approximate colloid content and the composition of the colloid were known in each case.

ADSORPTION OF SODIUM CARMINATE BY SOILS

The first acid dye used was sodium carminate. This, according to Way (*17*), is strongly adsorbed by clays. Solutions made of three different lots of sodium carminate purchased from a chemical supply house appeared to be highly but unequally adsorbed. However, a solution of one of these lots treated in a different manner was not adsorbed at all, and a fourth lot prepared from carminic acid and sodium hydrate likewise showed no adsorption at pH 7.2. Only a general description will be given of determinations made with the first lot of this dye, since there is doubt as to the reliability of the results as adsorption data.

Many determinations were made with the first stock solutions with a view to standardizing a method for comparing the adsorptions of different soils. It was found that decolorization of the dye solution was markedly affected by the time of shaking. Of the dye removed in 16 hours, about 70 per cent was taken out in the first 4 hours; but after 4 hours the rate of removal seemed to be practically constant up to 70 hours. Further, it was found that although the quantity of dye adsorbed showed some correspondence with the concentration of the dye solution, it was nearly a straight-line function of the quantity of dye supplied. These facts indicated that something besides an adsorption of an electrolyte was taking place. It was also found that duplicate determinations agreed well if made on the same day or a few days apart, but if made several weeks apart the duplicates varied widely. It then became apparent that the stock solution was deteriorating and that a precipitate was forming in the originally clear solution. Part of the precipitate was obviously due to a fungous growth, which Charles Thom of this bureau identified as a species of Aspergillus.

A few crystals of thymol added to a new stock solution seemed to be effective in keeping it free from fungous growth, but this solution also obviously changed on standing a few weeks.

Probably little significance should be attached to results obtained with these stock solutions since they deteriorated more or less and since subsequent lots of dye gave quite different results. However, such results as were obtained were only in very general accord with the idea that colloids with low silica-sesquioxide ratios would adsorb more of an acid dye than colloids with high silica ratios. The Sharkey soil, Norfolk subsoil, and Nipe soil, containing colloids with silica-sesquioxide ratios of 3.11, 1.60, and 0.31 removed, respectively, 0.074 0.303, and 0.489 g. of dye per gram of colloid; but four other soils containing colloids with ratios of 1.53 to 1.70 removed quantities of dye varying between 0.075 and 0.303 g. per gram of colloid.

A new stock solution was prepared in the same manner as the first two, except that the filtered dye solution was rendered slightly acid, shaken occasionally for three days, and again passed through a Pasteur-Chamberland filter. After removal of the precipitate by filtration, the solution was neutralized with dilute NaOH and then diluted to 0.2 per cent. The addition of acid to the dye was prompted by the idea that some of the dye in the first two solutions might have been in a finely divided colloidal state which passed through the filter but later coagulated and contributed to the gradual weakening of the dye solution. It was thought that if this were the case, acid would help to coagulate and remove an unstable part of the dye from the stock solution. This solution had less color than the preceding ones similarly acidified, owing either to mode of preparation or difference in the composition of the sample, although it presumably contained the same concentration of dye. This solution was only slightly adsorbed by the Sharkey soil and the Norfolk subsoil, and not at all by Nipe clay and three other soils.

A fourth lot of dye was then prepared from some supposedly pure carminic acid purchased from another chem cal supply house. Slightly less than the theoretical quantity of sodium hydroxide required to form sodium carminate was added to a solution of this carminic acid and the pH was brought to 7.2 by the addition of dilute sodium hydroxide. This dye solution was likewise not adsorbed by the Sharkey soil or the Norfolk subsoil until the pH was raised to 7.7, and then only slightly.

The failure of these soils to adsorb the acid-treated and prepared dye, except as noted, was assumed to be due to the kind of surface exposed by the colloid. It should be possible to change the nature of this surface very markedly by treatment with iron and aluminum salts. The two soils were therefore treated with ferric chloride and with aluminum chloride. Two lots of each soil were shaken for 17 hours with 5 per cent solutions of each chloride, in the proportion of 5 g. of soil to 100 c. c. of solution. The soils were then filtered and washed with distilled water. One half of each sample was washed twice, and the other half was washed repeatedly, about 15 times. After the soils had been dried at 105° C. and exposed to the air for a day or two, determinations were made of the adsorptions, as shown in Table 1.

TABLE 1.—*Adsorption of sodium carminate by soils treated with ferric and aluminum chlorides*

Soil	Treatment	Dye adsorbed per gram of colloid
		Gram
Norfolk fine sandy loam subsoil	$FeCl_3$, washed twice	0.248
	$FeCl_3$, washed repeatedly	.243
	$AlCl_3$, washed twice	.224
	$AlCl_3$, washed repeatedly	.219
Sharkey clay	$FeCl_3$, washed twice	.417
	$FeCl_3$, washed repeatedly	.383
	$AlCl_3$, washed twice	.300
	$AlCl_3$, washed repeatedly	.250

Evidently entrained soluble iron and aluminum chlorides played no appreciable part in the high adsorptions observed, for samples washed repeatedly removed almost as much dye as samples washed only twice. The high adsorptions may therefore be ascribed to changes in the colloids brought about by treatment with the salt solutions. These changes are not certain, but the following are to be expected: Some substitution of the exchangeable soil bases by iron and aluminum ions, and a union of the hydrous oxides or oxychlorides with the soil colloid particles. There is an indication in the comparative quantities of dye taken up by the two soils that removal of the dye was due chiefly to hydrous oxides precipitated on the surface of the colloids rather than to a precipitation of the dye anion by iron or aluminum ions released from the treated colloid by the sodium ions of the dye solution. If substituted iron or aluminum ions had been chiefly responsible for removal of the dye, it would seem that the Sharkey soil should have removed several times as much dye as the Norfolk, since its exchange base capacity is several times that of the Norfolk colloid (*2, p. 23*). As a matter of fact, the total quantity of dye removed by the four Sharkey samples is only 1.45 times the quantity removed by the four Norfolk samples. Curiously enough, the surface exposed by 1 g. of the Sharkey colloid is about 1.42 times that of the Norfolk colloid, as shown in a previous publication (*2, p. 4*).

There is thus some ground for assuming that treatment of the Sharkey and Norfolk soils with iron and aluminum chloride solutions introduced a reactive surface and that failure of the untreated soils to adsorb the sodium carminate to the same extent was due to lack of such a surface. The reactive surface introduced probably consisted of freshly precipitated gels of the hydrous oxides with more or less adsorbed chloride. As will be shown later, there is reason for believing that such a surface would lose reactiveness on prolonged washing and subsequent aging.

ADSORPTION OF ORANGE II AND PONCEAU BY SOILS AND ARTIFICIAL GELS

A number of other acid dyes were obtained from the color laboratory of the Bureau of Chemistry and Soils. These were tested qualitatively to see if they were suitable for adsorption determinations made by the colorimetric method. Since the soil suspensions

were clarified by the addition of a drop or two of acid and subsequent centrifuging, it was necessary to use a dye that was not precipitated by small amounts of acid and one that had a constant color in the presence of different degrees of acidity. Many dyes did not meet these requirements. Solutions of Congo red and metanil yellow, for instance, were almost completely precipitated by small amounts of acid. Ponceau and orange II, however, seemed to be satisfactory for this work.

These two dyes were used in adsorption determinations with several soils containing colloidal materials with widely different silica-sesquioxide ratios. Both water and N/2,000 hydrochloric acid solutions of the dyes were used. The hydrogen-ion concentrations of the dye solutions in contact with the different soils were determined with the hydrogen electrode. This determination is probably not very reliable in the presence of dye, but the pH values obtained for the soils in the unacidified dye solutions were about the same as for the soils in distilled water. The 0.1 per cent solution of ponceau in water had a pH of 7.2. The results obtained are shown in Table 2.

TABLE 2.—*The adsorption of orange II and ponceau in water and in N/2,000 HCl by soils containing colloids with different silica-sesquioxide ratios*

Soil	Orange II adsorbed per gram of colloid—		Ponceau adsorbed per gram of colloid—		SiO_2 / $Al_2O_3 + Fe_2O_3$ ratio of colloids	pH values of ponceau and soil—	
	In water	In N/2,000 HCl	In water	In N/2,000 HCl		In water	In N/2,000 HCl
	Mgm.	*Mgm.*	*Mgm.*	*Mgm.*		*pH*	*pH*
Sharkey clay, A horizon	20.0	33.0	0	0	3.07	6.3	5.9
Iredell loam, B horizon			3.0	5.0	1.88		
Norfolk fine sandy loam, B horizon	0	9.0	0	0	1.60	5.5	4.7
Davidson clay loam, B_1 horizon			9.0	19.0	1.50		
Cecil sandy loam, A_3 horizon	14.0	16.0	10.0	12.0	1.35	5.1	4.7
Nipe clay, A horizon	22.0	30.0	13.0	18.0	.31	7.1	7.2

The small adsorptions in water solutions are in strong contrast with the adsorptions obtained in previous work with the basic dye malachite green (*10, p. 11*). Whereas the six soils used in this work adsorbed from 0 to 22 mgm. of orange II and 0 to 13 mgm. of ponceau per gram of colloid, the 30 soil colloids used in the earlier work adsorbed from 77 to 431 mgm. of malachite green.

The small adsorptions of these acid dyes are, on the whole, related only slightly to the silica-sesquioxide ratios of the colloids. The four adsorptions of orange II show no correspondence to these ratios, and the lesser quantities of ponceau adsorbed show only a slight correspondence.

The use of N/2,000 hydrochloric acid increased the adsorption measurably; but the adsorption of these acid dyes was still small as compared with the adsorption of a basic dye. It should be noted, however, that owing to the neutralizing action of the soil, the use of this dilute acid increased the hydrogen-ion concentration of the solution only slightly.

Determinations were then made of adsorption from a much more acid solution by using N/20 hydrochloric acid solutions of the dyes. The results are given in Table 3.

TABLE 3.—*The adsorption of orange II and ponceau from N/20 hydrochloric acid solutions, and the quantities of ponceau precipitated by soluble salts extracted from the soils with N/20 acid*

Soil	Orange II adsorbed per gram of colloid	Ponceau adsorbed per gram of colloid	Ponceau precipitated by soluble salts extracted from soils with N/20 acid, calculated per gram of colloid
	Milligrams	*Milligrams*	*Milligrams*
Sharkey clay, A horizon	126	20	20
Iredell loam, B horizon		223	
Norfolk fine sandy loam, B horizon	64	23	23
Davidson clay loam, B_1 horizon		109	
Cecil sandy loam, A_2 horizon	64	40	27
Nipe clay, A horizon	208	104	42

It will be seen that the quantities of dye removed from solution were greatly increased in all cases by the twentieth-normal acid; but it did not seem likely that the large quantities of orange II and ponceau removed represented true adsorption. It is true the results can not be attributed to precipitation of the dyes by the acid of the medium, for preliminary tests showed the dyes were not precipitated by dilute acids. Furthermore, if removal of the dyes had been due to acid precipitation, the quantities of dye removed would have been similar for all soils, and this was far from being the case. There is, on the other hand, evidence that the high adsorptions were due, in part at least, to precipitation of the dye by iron, aluminum, and other bases released from the colloidal material by the acid used.

Qualitative tests showed that small quantities of the chlorides and sulphates of iron, aluminum, calcium, magnesium, manganese, and chromium precipitated 0.1 per cent solutions of orange II and ponceau to a greater or less extent. In a quantitative test with aluminum chloride, 0.003 gm. of this salt added to 40 c. c. of 0.1 per cent dye solutions precipitated 1.5 per cent of the ponceau and 31 per cent of the orange II.

Determinations were then made to see whether the bases released from the soils by N/20 acid would precipitate the quantities of dye reported as adsorbed in Table 3. Four soils were shaken 17 hours with N/20 hydrochloric acid; the proportions of soil to acid were kept the same as in the adsorption determination, but larger quantities were used. Two portions of the clear solution from each soil, corresponding to the soil weights of the adsorption determination, were evaporated to dryness on a steam bath. One series of the evaporated soil extracts was taken up with enough acid to give a N/20 solution on addition of the dye, and the other series was taken up with distilled water. The series of extracts dissolved in acid gave no precipitates

with ponceau; but the extracts dissolved in water precipitated the quantities of dye reported in column 4 of Table 3.

The fact that the extracted salts dissolved in water did precipitate the dye, whereas the same salts dissolved in N/20 acid did not, shows that the precipitation is markedly influenced by acidity. In view of this influence of acidity, it is impossible to tell from the data given in Table 3 just how much dye was precipitated in the adsorption determinations, since the solution of extracted salts in water was obviously less acid than the solution of dye in N/20 acid used in the adsorption determination. However, the data strongly support the idea that most of the dye removed from the acid-dye solution was by precipitation with dissolved bases rather than by adsorption. This applies also to the sodium carminate in acid solution, some of which, however, was precipitated directly by the acid.

Since even soils with low silica-sesquioxide ratios adsorbed only small quantities of ponceau or orange II from water solutions, tests were conducted with artificial gels in order to see if they would take up appreciable quantities of these acid dyes. The gels used comprised the following:

(1) A commercial alumina gel that was in a hard granular condition. This gave off no water on heating to 105° C., but on ignition it lost 34.97 per cent of water as compared with the 34.57 per cent of water present in aluminum hydroxide.

(2) An air-dried alumina gel prepared in this laboratory. It contained 0.25 per cent chlorides, and lost 21.8 per cent water on drying at 105° C. and 40.35 per cent water on ignition.

(3) A commercial iron gel in a pasty condition. This lost 85.55 per cent water on drying at 110° C. and 92.85 per cent on ignition.

(4) An air-dried iron gel prepared in this laboratory. This contained 0.09 per cent chlorides and lost 3.8 per cent water on drying at 110° C. and 8.3 per cent water on ignition.

The iron and alumina gels prepared in this laboratory were made by G. J. Hough by precipitating aluminum chloride or ferric chloride with ammonia. They were washed repeatedly by the use of Pasteur-Chamberland filters and suction until the wash waters showed only traces of chlorides. Prior to use in the adsorption determinations they had been kept a few weeks in the air-dried condition. The quantities of ponceau adsorbed by these gels before and after heating are shown in Table 4.

TABLE 4.—*The adsorption of ponceau by artificial gels of the hydrous oxides of iron and aluminum*

Kind of gel	Dye adsorbed per gram of gel dried at 105° or 110° C.	Kind of gel	Dye adsorbed per gram of gel dried at 105° or 110° C.
	Milligrams		*Milligrams*
Commercial alumina gel	0	Commercial iron gel	228
Commercial alumina gel, dried at 110° C.	0	Commercial iron gel, dried at 110° C.	71
Commercial alumina gel, dried at 600° C.	0	Commercial iron gel, dried at 600° C.	14
Prepared alumina gel, air dried	67	Prepared iron gel, air dried	13
Prepared alumina gel, dried at 105°C.	13	Prepared iron gel, dried at 110° C.	13
Prepared alumina gel, ignited strongly by blast	0	Prepared iron gel, ignited strongly by blast	0

The failure of the commercial alumina gel to adsorb ponceau and the low adsorption of the prepared iron gel are similar to the results observed with soils. The other two gels, however, had comparatively high adsorption capacities. These results can be explained somewhat more satisfactorily than the results obtained with different soils, since previous work with the simpler, artificial gels has shown something of the characteristics of these materials affecting the adsorption of electrolytes.

It is well known that the adsorptive capacities of the hydrous oxide gels are affected by their previous history and manner of preparation. Old gels, gels that have been dried at high temperatures, and gels prepared especially free of electrolytes are likely to be poor adsorbents of electrolytes. These characteristics of age, temperature at which the gel is dried, and content of electrolytes can be used, of course, only in a general way for predicting the adsorptive capacity of a gel for electrolytes, since one condition probably influences the effect of another. It seems probable, for instance, that the loss of adsorptive capacity on aging is markedly affected by the electrolyte content, aging being more rapid when the electrolyte content is low. This is suggested by Weiser's recent work with an alumina gel free from electrolytes, prepared from amalgamated aluminum (*18*). This gel adsorbed alizarin when freshly prepared, but lost practically all adsorptive power in a few days.

The alumina and iron gels which adsorbed comparatively large amounts of ponceau lost the greater part of their adsorptive capacities when dried at 110° C. The failure of the commercial alumina gel to adsorb may be attributed to a possible drying at high temperature or to aging. This preparation, which was hard and granular, gave off no water at 105°, although on ignition it gave off almost the correct amount of water for crystalline aluminum hydroxide. The difference in the adsorptive capacities of the prepared alumina and iron gels may be attributed partly to a difference in purity, the alumina gel containing 0.25 per cent chloride and the iron gel 0.09 per cent. The high adsorption of the commercial iron gel is probably to be similarly explained.

These results with alumina and iron gels tend to confirm those obtained by several other investigators previously cited in showing that all extended iron and aluminous surfaces do not adsorb acid dyes. Adsorption evidently takes place only when these surfaces are in a reactive condition.

DISCUSSION OF RESULTS

The results obtained with these three acid dyes, sodium carminate (prepared from carminic acid), orange II, and ponceau, indicate that most soils have very small or almost no adsorptive capacities for acid-dye anions. Even soils containing colloids with low silica-sesquioxide ratios adsorbed very little. The comparatively high adsorptive capacity of charcoal for both acid and basic dyes, observed by Michaelis and Rona (*13*), suggests that possibly soils high in organic matter might adsorb more than the soils used in this investigation.

This conclusion of the low adsorbability of acid-dye anions, based on work with three acid dyes, is in harmony with the conclusions of various investigators that adsorption of the simpler anions Cl, NO_3,

and SO_4 by soils is negligible in neutral solutions. Some of the previous work on dye adsorption by soils also indicates a negligible adsorption of acid-dye anions; but the results of other investigators are apparently contradictory to this conclusion.

It is of course evident from the results of previous investigators that soils may in some cases take up appreciable quantities of certain acid dyes. But it is questionable whether in these cases the removal of the dye is a true adsorption of anions. The results obtained in this study and the known characteristics of dye solutions suggest that the high adsorptions previously reported may have been due to removal of dye colloids, to precipitation by acid, or to precipitation by bases in solution, rather than to adsorption.

It is well recognized that dyes, with their large molecules, tend to form colloidal solutions; some dyes, as Congo red and night blue, seem to be distinctly colloidal; others are what Freundlich (*9, p. 893*) terms "halbkolloide"; and some dyes, as eosin and methylene blue, are dispersed chiefly as electrolytes. A finely divided suspension of the more colloidal dyes might well be taken up by the only slightly dispersed soil colloids on prolonged shaking when anions would not be adsorbed. Michaelis and Rona (*13*), for instance, noted a removal of the colloidal Congo red by kaolin but no adsorption of potassium eosinate. The results obtained in this work with the first two lots of sodium carminate probably were also due in part to a removal of dye colloids. Furthermore, many acid dyes are precipitated or coagulated by slight concentrations of acid and by small quantities of the salts of calcium, magnesium, iron, aluminum, manganese, or other bases. In some cases the precipitate formed may become apparent only on centrifuging. Some high adsorptions of acid dyes previously reported might well have been precipitations of the dye with bases released from the soil colloids by base exchange with cations present in the dye preparations. The large amounts of metanil yellow that Marker and Gordon (*12*) report as adsorbed by silica, alumina, and iron gels were probably simple precipitations, by acid alone in the silica gel test and by acid or dissolved iron and aluminum in the tests with alumina and iron gels.

The idea that soil colloids might be better characterized by the comparative quantities of acid and basic dyes adsorbed than by the single adsorption of a basic dye or inorganic cation is apparently not well founded. The results of this study indicate that the quantity of acid dye taken up is either too small to be significant or is open to question as being true adsorption.

The somewhat negative results of this investigation are, however, suggestive of certain features of the surface exposed by soil colloids. The small adsorptions reported bear little relation to the comparative quantities of sesquioxides in the colloid and probably they afford little indication of the comparative amounts of iron and aluminum present in the surface either as oxides or silicates. A failure to adsorb simple anions and the complex acid-dye anions might, of course, be attributed to a lack of iron and aluminum radicals in the surfaces exposed to solutions; but it seems more reasonable to attribute this lack of adsorption to an unreactive condition of these basic radicals, if work on artificial iron and alumina gels is considered.

Data given in Table 4 and the results of other investigators show that some alumina and iron gels are active adsorbents, whereas others are not. The adsorptive gels seem to be those that are freshly prepared and those that contain an ionic impurity, either adsorbed or retained by the gel during its preparation. These facts might be interpreted in the case of an alumina gel, for example, by assuming that a pure, freshly prepared alumina gel contains some exchangeable hydroxyl ions, while an impure gel contains some reactive aluminum chloride, sulphate, or nitrate; that on aging of the pure gel or on removal of the anion from the impure gel, these reactive materials tend to pass into an unreactive hydrous oxide surface. The observed changes in the structure of gels that take place on aging might also be dependent to some extent on such hypothetical chemical changes.

The preceding facts and hypotheses may not apply to soil colloids, for the soil colloidal material is certainly more complex than an artificial hydrous oxide gel. But on the assumption that they do apply, the following hypotheses are suggested: Soil colloids being thoroughly aged, an adsorption of anions would not be expected unless adsorbed anions were already present. The low adsorptive capacity for acid-dye anions, therefore, indicates that the iron and aluminous surfaces carry few, if any, adsorbed anions. This may be due to such surfaces being present in an unreactive condition as highly insoluble hydrous oxides. Such surfaces might be rendered reactive to anions by solutions sufficiently acid to permit some ionization.

SUMMARY

The fact that the adsorbed cations of the basic dyes by soil colloids vary roughly with the proportion of silica to iron and aluminum in the colloid suggests that a similar relationship might obtain for the adsorption of acid-dye anions. Although adsorption of the simple anions from neutral solutions seems to be negligible, several investigators have reported appreciable adsorptions of dye anions of the acid dyes.

Data are given on the adsorption of the anions from water and weakly acid solutions of the acid dyes, sodium carminate, orange II, and ponceau. Several soils containing colloids with widely different silica-sesquioxide ratios adsorbed these dyes slightly or not at all from water solutions. Adsorptions from N/2,000 acid solutions of the dyes were larger but also inappreciable. From the N/20 acid solution large quantities of ponceau were removed by some soils; but evidence is given that removal of the dye in this case was a precipitation of the dye with bases dissolved from the soil colloids rather than a true adsorption.

It is concluded that the true adsorption of anions of the acid dyes by soil colloids is negligible, like that of the simpler anions. The comparatively large adsorptions of acid dyes reported by several earlier investigators are ascribed to the use of colloidal dyes and to precipitations of the dyes by acid or by bases released from the soil colloids.

The apparent lack of adsorptive capacity for anions is discussed in its relation to the nature of the surface exposed by soil colloids.

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THE RELATIVE RESISTANCE OF VARIETIES AND SPECIES OF CITRUS TO PYTHIACYSTIS GUMMOSIS AND OTHER BARK DISEASES[1]

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INTRODUCTION

Although the great importance of knowing the response of the varieties and species of Citrus to various pathogenes is at once apparent, records of complete and thoroughgoing tests and surveys are not numerous. The dearth of data acquired by artificial inoculation of the experimental hosts under more or less accurately controlled or comparable conditions is in part understandable when one considers the general feeling among growers that such inoculations may result in injury and possible death of valuable plants. Also there is the difficulty of obtaining the many varieties under uniform conditions of age, development, and health.

REVIEW OF LITERATURE

The work of Lee (*5*, *6*[2]), Peltier (*9*), and Peltier and Frederich (*10*, *11*) on the susceptibility and resistance of various rutaceous hosts to citrus canker is noteworthy from the standpoint of the number and diversity of plants tested and observed. In descending order of susceptibility stand pummelo, trifoliate orange, the various limes, sweet orange and lemons, sour orange, varieties of the mandarin group, calamondins, citrons and kumquats, the last four being considered commercially resistant. Lee has found the Triumph variety of pummelo to exhibit a slight resistance to the bacterium, and he classifies also Tahiti lime as commercially resistant. McLean (*7*) and McLean and Lee (*8*) noted a correlation between the width of the stomatal slit and susceptibility to canker, their explanation being that the narrow stomata offer less easy ingress to water which may be laden with the causal organism.

Fulton (*2*) in his extensive investigation on the susceptibility of Citrus varieties to the West Indian lime anthracnose organism, *Gloeosporium limetticolum* Clausen, showed that only the West Indian and Dominican thornless varieties of lime are very susceptible, six other varieties of lime giving no positive indication of susceptibility either in the greenhouse or in the field. Certain strains of citron when wounded and kept under greenhouse conditions became infected to a very limited extent. All other genera, species, varieties, and hybrids of the Rutaceae tested were either immune to the disease or could not be unquestionably infected. On the other hand the anthracnose or withertip fungus, *Colletotrichum gloeosporioides* Penz., does not limit

[1] Received for publication June 2, 1930, issued September, 1930. Paper No. 222, Univeristy of California, Graduate School of Tropical Agriculture and Citrus Experiment Station, Riverside, Calif. This is the third of a series of papers (*3*, *4*) on the parasitism of *Phytophthora citrophthora* (Sm. and Sm.) Leonian.

[2] Reference is made by number (italic) to Literature Cited, p. 424.

its attack to a single species but parasitizes the mature twigs and fruit of many species (*1, p. 287–293*).

A third citrus disease on which extensive tests have been made is verrucosis or scab caused by *Sphaceloma fawcettii* Jenkins. Winston (*12*) and Winston, Bowman, and Bach (*13*) have obtained results which enabled them to separate the various rutaceous hosts into four catagories based on their susceptibility, resistance, and immunity to the disease. Among the better known varieties and species, sour orange and lemon were severely attacked, the tangerine and some pummelos moderately attacked, sweet orange and Tahiti lime only rarely parasitized, while citron, kumquat, and the Royal and Triumph varieties of pummelo were not attacked. In his 1923 publication Winston (*12*) records that the fruit of pummelo is susceptible to scab only from its earliest formation up until it has reached three-fourths of an inch in diameter, after which it becomes immune. Similarly, leaves are immune after they reach one-half inch in width. With Pythiacystis rot the writers have observed a relationship approximating the opposite of that with scab, namely, in the case of twigs, branches, and leaves, the more mature the organ the greater is its susceptibility. While all ages and sizes of fruit are susceptible to Pythiacystis brown rot, immature fruit decays less rapidly than ripe fruit. The investigagations referred to above and the data presented in this paper, the latter being a record of tests made by the writers during the years 1926, 1927, and 1928, constitute the more extensive contributions to the study of disease resistance and susceptibility in Citrus.

Records of observations on the response of the more important commercial members of the genus Citrus to bark diseases cover a period of years approximately concurrent with the commercial production of citrus fruit. As a result it is now definitely known generally that lemon, orange, and pummelo are susceptible to Pythiacystis gummosis and that sour orange is resistant; that the sweet orange and pummelo are susceptible to psorosis; and that the lemon stands with sour orange in resistance. To decorticosis the citrus hosts react still differently, lemon alone being susceptible, and sweet orange, pummelo, and sour orange completely resistant or immune. Since with Pythiacystis gummosis a good indication of the reaction of a given species or variety of Citrus may usually be obtained in 40 days, whereas with psorosis and decorticosis several years are required even on the more susceptible hosts, the experimental data reported here are confined to the first disease. For purposes of comparison it may be recalled from previous discussion that sour orange, lemon, and pummelo are susceptible to scab, while the sweet orange is commercially resistant. All four species, however, are found to be susceptible to canker. These comparisons of the very variable reactions of the four important commercial species of Citrus indicate that we should look to the specific biochemical make-up of the protoplasm for hypotheses that may lead to explanation.

MATERIALS AND METHODS

The inoculation tests on brown-rot gummosis reported here were made in the variety orchards of the University of California Citrus Experiment Station. Subcultures of a strain of *Phytophthora citrophthora* (Sm. and Sm.) Leonian, known as 1309–a, which was isolated from a diseased lemon tree at La Habra, Calif., were used throughout

the work. The surfaces of the places on the tree trunk to be inoculated were in all cases first rubbed with a towel dipped in 95 per cent alcohol. The 1926 inoculations were confined to 9-year-old trees of the Rubidoux plot, and were made during the period January 2 to 5. A small piece of mycelium from a 6-day-old glucose-potato agar slant culture was inserted under the flap of bark made by a small vertical slit. The inoculations were made on the east or southeast side of the trunk and on one main limb and were covered with pieces of oiled paper, the edges of which were glued. The 1927 inoculations were made during the period February 22 to March 4 in fields 1 and 12. Each tree received one inoculation on the west side of its trunk in the manner described.

Inoculations in 1928 were made during March 15 to March 26 in all three of the above-mentioned groves. In general, depending upon the size of the tree, four inoculations, two about 6 inches and two about a foot from the ground, were made on the trunk of each tree, the fungus being inserted at each of the four points of the compass. Instead of a vertical slit a small cylindrical hole, 4 mm. in diameter, was drilled through the bark to the cambium with a flamed cork borer. The inoculum was placed directly on the cambium and the hole sealed with a piece of adhesive tape, the portion directly over the inoculum being lined with a half-inch square piece of paper to prevent fungicidal action by the zinc oxide of the tape.

There were three sets of observations and measurements. At the first inspection, 40 to 45 days after inoculation, only external length and breadth of the lesion were measured, the disease being allowed to run its course undisturbed until 20 days later when similar external measurements were made. The canker was then excised, and measurements were taken of the lesion as it appeared on the surface of the cambium and wood. Finally the area of the lesion on the excised bark was measured by means of a planimeter.

TABLE 1.—*Size of lesions in square centimeters made and amount of gum formed on 11-year-old citrus trees by Phytophthora citrophthora in 60 days*

[Field No. 1. Inoculations made March 22, 1928; excisions made May 22, 1928]

Species and variety	Experiment station No.	Lesion measurements						Order of resistance	Amount of gum exuding[a]
		North	South	East	West	Individual average	Group average		
Lemon (Citrus limonia):									
Seedling	612			28.4	21.1	24.8		1	
Villafranca	568			33.3	21.9	27.6		2	
Do	280			29.0	38.7	33.8		3	
Kennedy	244			36.0	36.6	36.3	35.3	4	
Villafranca	390			29.2	50.6	39.9		5	
Eureka	583			56.75	25.21	41.0		6	
Belair	566			[b] 26.60	60.6	43.6		7	5
Rough	400	7.55	4.06	12.26	3.42	6.82	6.82	------	3
Lime (Citrus aurantifolia):									
Tahiti	391	1.935	1.55	.3875	------	1.29		1	1
Sweet (lemon), Florida	569	3.36	4.06	------	------	3.71		2	1
Kusia	452	3.226	5.8	3.42	------	4.15	4.02	3	1
Sweet	654	4.52	4.06	4.06	------	4.2		4	2
Do	654	5.48	3.875	------	------	4.68		5	3
Rangpur	131	8.20	4.13	5.16	6.9	6.1		6	2
Citron (Citrus medica):									
Citron of commerce	649	------	------	[c] 1.78	[d] 1.162	1.47	1.54	1	1
Indian	138	------	------	1.613	1.613	1.613		2	1

See footnotes at end of table.

TABLE 1.—*Size of lesions in square centimeters made and amount of gum formed on 11-year-old citrus trees by Phytophthora citrophthora in 60 days*—Continued

[Field No. 1. Inoculations made March 22, 1928; excisions made May 22, 1928]

Species and variety	Experiment station No.	Lesion measurements						Order of resistance	Amount of gum exuding[a]
		North	South	East	West	Individual average	Group average		
Pummelo (Citrus maxima):									
Siamese	640	2.39	1.29	1.613	2.32	1.903		1	1
Round Hawaiian	448	1.162	1.935	3.87	.....	2.32		2	1
Imperial	596	3.74	5.94	3.42	6.58	4.92		3	2
Windsor	350	4.0	6.452	4.91	5.16	5.13		4	3
Marsh	126	4.52	6.13	6.13	4.96	5.43		5	3
Pernambuco	298	1.484	6.65	5.81	8.52	5.62	6.505	6	2
Chinese	325	6.58	.....	[b]4.77	[c]5.55	5.63		7	1
Hall Silver	256	8.39	5.8	7.81	4.26	6.56		8	3
Duncan (path of infection at 45° angle)	246	6.452	11.86	8.26	21.93	12.13		9	5
McCarty	265	26.5	18.25	7.87	9.04	15.4		10	3
Sweet orange (Citrus sinensis):									
Navel	397	3.42	4.0	3.61	4.06	3.77		1	1
Indian River	355	4.06	4.52	4.26	3.68	4.13		2	1
Catlin	429	5.29	3.68	4.84	5.62	4.85		3	1
Boone Early	260	4.71	2.71	6.452	5.08	4.89		4	1
Weldon	430	3.23	6.71	7.43	4.71	5.52		5	1
Hart Tardiff	570	5.62	4.84	7.29	6.65	6.1	6.08	6	2
Lue Gim Gong	255	7.74	5.16	5.36	6.452	6.18		7	2
Parson Brown	295	.....	7.94	4.45	7.68	6.69		8	1
Madam Vinous	234	7.94	8.20	4.91	6.77	6.95		9	1
Selecta	37782	9.68	8.71	6.00	6.452	7.71		10	1
Sweet Seville	286	8.20	11.42	12.90	8.13	10.16		11	3
Mandarin (Citrus nobilis):									
Satsuma	592	.3226	1.033	.903	.....	.753		1	±
Cleopatra	270	1.484	1.42	1.42	1.613	1.484		2	1
Willow Leaf	595	4.71	4.0	4.65	2.49	3.96	4.15	3	0
Oneco	263	5.62	3.94	5.03	4.90	4.87		4	1
Clementine	279	4.38	5.49	7.75	7.23	6.21		5	1
Dancy	593	8.13	7.94	8.9	5.62	7.65		6	2
Sour Orange (Citrus autantium):									
Seedling[f]	.....	.....	.....	.774	.6452	.71		1	±
Do.[f]	.....	.....	.....	1.42	.9675	1.194	1.368	2	±
Paraguay	660	1.68	1.74	1.68	.....	1.7		3	±
Standard Seedling	628	1.68	2.13	2.066	1.613	1.87		4	±
African[f]	64	[g]	[g]	[g]	[g]	[g]	[g]	5	.....
Kumquat (Fortunella spp.):									
Marmumi	132	.904	1.097	.....	1.355	1.119	1.204	1	1
Nagami	268	1.29	.....	.....	.....	1.29		2	1
Calamondin (Citrus mitis):[f]									
Calamondin	643	.....	.....	1.806	1.42	1.613	1.806	1	1
Do	643	3.36	.....	.6452	.....	2.00		2	1
Other species:									
Citrus ichangensis[f]	1215	2.58	4.71	3.1	2.26	3.16	3.61	1	3
Do.[f]	1215	7.93	.....	2.26	2.00	4.06		2	3
Citrus hystrix[f]	432	.....	.....	.968	1.29	1.129	1.63	1	1
Do.[f]	432	2.26	.....	2.45	1.68	2.13		2	1
Citrus limetta[f]	758	[h]	.....	[h]	.....	.....		.....	±
Citrus webberii[f]	767	.....	.....	6.71	3.61	5.16	22.78	1	4
Do.[f]	767	.....	.....	54.24	26.65	40.4		2	5
Hybrids:									
Tangelo (Tangerine×Pummelo)—									
Sampson	399	1.613	1.613	1.613	2.77	1.902	1.905	1	±
Citrange (Trifoliate×Sweet)—									
Cunningham	271	.6452	4.84	.1935	2.49	2.02		1	1
Sanford	276	2.9	4.0	.516	.....	2.47		2	1
Savage	275	3.35	2.84	3.29	.....	3.16	12.75	3	1
Morton	296	2.77	24.9	4.52	8.38	10.1		4	1
Rusk	301	6.20	46.25	18.25	.....	23.57		5	5
Eatonia	309	8.39	78.07	.....	19.0	35.15		6	5
Colman	250	[i]	[i]	[i]	[i]	[i]	[i]	7	5

[a] Expressed on a numerical basis, 5 representing a copious secretion, 4 medium plus, 3 medium, 2 medium minus, 1 slight, and 0 none.

[b] Southeast.

[c] Northeast.

[d] Northwest.

[e] Southwest.

[f] Inoculation made May 4, 1928, excision made June 11, 1928; 38 days.

[g] No involvement. Immune.

[h] Slight formation of gum.

[i] Girdled. Very severe. Could not excise and save bark.

TABLE 2.—*Average areas of lesions in square centimeters made on citrus trees by Phytophthora citrophthora and amount of gum exuding*

Species and variety	Average of lesion measurements after—						Grand average for the variety	Grand average for the species	Amount of gum exuding[a]	Average amount of gum exuding
	67 days on 9-year trees, Rubidoux plot, 1926	110 days on 2½ to 3 year trees, field No. 12, 1927	74 days on 10-year trees, field No. 1, 1927	43 days on 3¼ to 4 year trees, field No. 12, 1928	70 days on 11-year trees, Rubidoux plot, 1928	60 days on 11-year trees, field No. 1, 1928				
Lemon (Citrus limonia):										
Ponderosa			16.25				16.25		+4.0	
Villafranca	42.0		60.00			33.8	45.3		+4.8	
Belair						43.6	43.6		+5.0	
Eureka	40.25		58.5			41.0	46.58	53.76	+4.4	
Seedling			85.6			24.8	55.2		+4.5	
Lisbon	29.1		125.0				77.0		+5.0	
Kennedy			148.5			36.3	92.4		+5.0	[b]4.67
Rough	11.0	13.12	23.87	4.64	3.9	6.82	10.56	10.56	+1.33	
Lime (Citrus aurantifolia):										
Bearse Seedless	[c]	[c]	[c]	[c]	[c]	[c]	[c]	[c]	[c]	
Kusia			9.0			4.15	6.57		+1.0	
Sweet (lemon)			10.0		9.03	3.71	7.58		+2.33	
Sweet			9.0		6.24	4.44	6.56	12.97	+2.5	
Tahiti	17.5		13.5			1.29	10.76		+2.0	
Rangpur	22.5		38.5			6.1	22.37		+2.66	
Persian Sweet	24.0						24.0		+3.0	1.93
Citron (Citrus medica):										
Italian	[d]	[d]	[d]						+1.0	
Indian						1.613	1.613	5.701	+1.0	
Chinese			7.25				7.25		+2.0	
Citron of Commerce	5.25		18.0			1.47	8.24		+1.66	1.41
Pummelo (Citrus maxima):										
Siamese						1.903	1.903		+1.0	
P. J. Wester seedling			3.75				3.75		0.0	
Imperial						4.92	4.92		+3.0	
Windsor						5.13	5.13		+3.0	
Red Fleshed					7.37		7.37		+3.5	
Marsh	18.0		8.00		6.83	5.43	9.56		+3.0	
Triumph	4.5		13.5		7.64		8.55		+4.0	
Hall Silver Cluster	13.5		8.75		6.29	6.56	8.78	10.54	+3.0	
Florida seedling	10.0						10.0		+2.0	
McCarty					4.97	15.4	10.18		+3.5	
Duncan	12.0				7.162	12.13	10.43		+4.0	
Round Hawaiian			25.0		4.805	2.32	10.71		+2.3	
Chinese			25.5			5.63	15.56		+1.0	
Pernambuco	5.25				32.53	5.62	14.47		+3.3	
Alonzo	17.5						17.5		+3.0	
Royal	8.0				51.7		29.85		+4.0	2.72
Sweet orange (Citrus sinensis):										
Golden Buckeye Navel	[c]	[c]	[c]						+3.0	
Indian River	3.0				3.645	4.13	3.59		+1.3	
Jaffa	4.5						4.5		+3.0	
Boone Early					5.34	4.89	5.11		+1.0	
Brazilian	5.0						5.0		+1.0	
Lue Gim Gong	4.5		8.0			6.18	6.23		+2.0	
Enterprise Seedless					6.56		6.56		+2.0	
Valencia	13.1	4.8		2.88			6.93		+1.0	
Sweet Seville	8.0				3.68	10.16	7.28	8.08	+1.3	
Dugat	8.0						8.0		+1.0	
Parson Brown			10.0			6.69	8.34		+1.0	
Pineapple			9.0		9.28		9.14		+2.0	
Homosassa			10.0		8.84		9.42		+1.5	
Ruby Blood	10.0						10.0		+1.0	
Navel	7.0	20.56	38.0	5.667	12.34	3.77	14.55		+2.0	
Mediterranean Sweet	12.0		21.0				16.5		+2.5	1.67

[a] Expressed on a numerical basis, 5 representing a copious secretion, 4 medium plus, 3 medium, 2 medium minus, and 1 slight.
[b] Rough omitted.
[c] Slight brown stain. Healed.
[d] Flap only killed. Healed.
[e] Very slight spread. Callus.

TABLE 2.—*Average areas of lesions in square centimeters made on citrus trees by Phytophthora citrophthora and amount of gum exuding*—Continued

Species and variety	Average of lesion measurements after—						Grand average for the variety	Grand average for the species	Amount of gum exuding[a]	Average amount of gum exuding
	67 days on 9-year trees, Rubidoux plot, 1926	110 days on 2½ to 3 year trees, field No. 12, 1927	74 days on 10-year trees, field No. 1, 1927	43 days on 3¼ to 4 year trees, field No. 12, 1928	70 days on 11-year trees, Rubidoux plot, 1928	60 days on 11-year trees, field No. 1, 1928				
Mandarin, (Citrus nobilis):										
Satsuma	1.125		5.25			1.04	2.47		+.5	
Willow Leaf						3.96	3.96		0.0	
Cleopatra	6.0		6.0			1.484	4.495		+.5	
Trimble			5.0				5.0		+3.0	
Parson Special	8.0		2.5				5.25		0.0	
Oneco			7.0		5.75	4.87	5.87	7.634	+1.0	
Clementine	10.0		7.0		7.66	6.21	7.72		+1.25	
Weshart			8.0				8.0		+3.0	
King	15.0		6.0				0.5		+1.0	
Dancy	33.0		8.8		4.935	7.65	13.6		+1.5	
Lester	40.0		5.44		5.89		17.11		0.0	1.07
Sour Orange (Citrus aurantium):										
Stock				1.161			1.161		+1.0	
Paraguay			3.9			1.7	2.8		+0.5	
Seedling			5.0			1.25	3.12	4.52	0.0	
Standard			7.5		2.55		5.02		+1.0	
Florida	7.0						7.0		+1.0	
South African	8.0						8.0		+1.0	.75
Trifoliate (Poncirus trifoliata):										
Trifoliate Stock No. 12							0.0		+1.0	
Do				.355			.355	.355	+1.0	
Trifoliate Rubidoux (Identity Questionable)					356.25		356.25	356.25	+5.0	2.3
Citrus limetta									±	
Microcitrus, (Microcitrus virgata)		2.7		1.27			1.99	1.99	+1.0	
Citrus hystrix			3.0			1.63	2.31	2.31	+1.0	
Citrus webberii			16.0			22.78	19.39	19.39	+4.0	
Citrus ichangensis						3.61	3.61	3.61	+3.0	
Calamondin, (Citrus mitis)			3.0			1.806	2.403	2.403	+1.0	
Severinia, (Severinia buxifolia)				.387			.387	.387	+2.0	
Kumquat (Fortunella):										
Marmumi						1.119	1.119		+1.0	
Nagami						1.29	1.29	1.205	+1.0	1.0
Tangelo, (Tangerine×Pummelo), Sampson	12.0		2.28		3.824	1.902	5.0	5.0	+1.0	
Citrange (Trifoliate×Sweet):										
Sanford			5.0		1.13	2.47	2.87		+.5	
Cunningham			12.5		.97	2.04	5.17		+.75	
Savage			36.0			3.16	19.58		+2.0	
Rusk	44.0	4.52	7.0	1.51	40.52	23.57	20.19	47.72	+2.4	
Morton	22.0		82.8	2.06	5.29	10.1	24.45		+2.3	
Eatonia			42.0		6.26	35.15	27.80		+3.25	
Colman			234.0				234.0		+3.3	2.07
Shaddock×St. Michael			3.0				3.0	3.0	+4.0	
Orange×Lemon			26.25				26.25	26.25	+3.0	
Citrangequat (Citrange×Kumquat)		2.87		.1548			1.51	1.51	+1.0	
Citrumelo (Trifoliate×Pummelo)				1.017			1.017	1.017	+2.5	
Citrangor (Citrange×Sweet)				2.192			2.192	2.192	±	
Limequat (Lime×Kumquat)		3.6		.605			2.102	2.102	+2.0	
Calamelo (Calamondin×Pummelo)		21.0		.908			10.984	10.984	+3.0	
Citruvel (Navel×Trifoliate)				.613			.613	.613	±	
Cicitrange (Trifoliate×Citrange)				1.162			1.162	1.162	+1.0	

[a] Expressed on a numerical basis, 5 representing a copious secretion, 4 medium plus, 3 medium, 2 medium minus, and 1 slight.

DISCUSSION OF RESULTS

Table 1 gives in detail the planimeter measurements of lesions obtained in 1928 in field 1, which contain the most representative and comparable lot of trees. The various species and varieties are grouped for purposes of comparison. Table 2 is a general summary of all the inoculation data of the three years, all the lesions on all the individuals of a variety being averaged. The data shown in the second, third, and fourth columns of Table 2, which give the results of the 1926 and 1927 investigations, were not obtained by planimeter measurements but by multiplying the length of the lesion by its breadth, inasmuch as the diseased areas were irregular in shape this method of calculation yielded values from 25 to 50 per cent greater than the actual values. However, the values of the respective groups thus estimated are reasonably comparable and are therefore included.

Since the trees in field 12 were only 3 to 4 years of age and since the writers have found that the younger organs of a lemon tree are less susceptible to Pythiacystis gummosis than the older, the results of the inoculations in that plot are not comparable with those obtained in the Rubidoux plot and in field 1 on trees 10 years old. The group results from the Rubidoux plot and field 1 approximated each other, and for simplicity there are presented in detail only the 1928 results in field 1, as recorded in Table 1. The relation of different groups in the other orchards was found in general to be the same.

A number of important conclusions appear to be justified by the data obtained. The varieties of lemon are uniformly very susceptible, only the seedling tree giving an indication of lessened susceptibility, and suggesting but a very remote possibility of obtaining by selection a variety of lemon resistant to the parasite. The long-observed and acclaimed resistance of sour orange has been substantiated. Here again the seedling form of the variety shows a greater resistance than the budded, in fact so great a resistance that for all practical purposes it may be considered to be immune. However, one specimen of the African variety of sour orange showed no involvement of tissue, which also suggests a true immunity.

Considering further the commercial varieties the writers found that grapefruit and sweet orange were about equal in susceptibility. As a group the varieties of *Citrus maxima* exhibit great variability of response to the parasite, ranging from a resistance only slightly less than that of the standard sour orange in the case of the Siamese variety to a susceptibility approaching lemon in the McCarty variety. In fact, the pummelo variety Royal Bahaman in the Rubidoux plot exceeded the most susceptible lemon as shown by an average involvement of 51.7 sq. cm. A peculiarity noted in the McCarty, the Royal, the Pernambuco, and in several other varieties of pummelo was that the disease progressed approximately at a 45° angle to the vertical. For this reason infection in these species presents a very serious aspect, that of girdling the tree possibly in a single season.

The variability in the varieties of sweet orange was less than in pummelo, and none approached the susceptibility of lemon. The average of the mandarin group was much below that of pummelo and sweet orange, being similar in this respect to rough lemon and the limes, *Citrus aurantifolia*. The Willow Leaf and Cleopatra varieties of mandarin showed considerable resistance in these tests.

According to the writers' observations one specimen of Tahiti lime could be classed with sour orange. Although general observations are that citron is very susceptible to gummosis, the two varieties inoculated here showed pronounced resistance to the strain of fungus employed. However, these two specimens of citron were apparently in a poor, nonsucculent condition, which may account for their lack of susceptibility. The evidence appears sufficient to warrant the trial of some of the unusual varieties as rootstocks, such as calamondin, and some of the hybrids—tangelo and the Cunningham, Sanford, and Savage citranges. Specimens of the Rusk citrange were very susceptible, as were also the Colman and Eatonia. The genus Fortunella, the kumquats, exhibited great resistance to the fungus, but because of their dwarflike growth their use as rootstocks is probably out of consideration. *C. limetta* and *C. hystrix* were resistant, *C. ichangesis* was less so, while one specimen of *C. webberii* succumbed as readily as the lemon varieties.

The amount of gum formed was approximately directly proportional to the extent of involvement of the disease. Gumming is the most prominent symptom of this disease, and on individuals of the several commercial kinds of Citrus one may obtain an approximate indication of the severity and extent of the malady by observation of the amount of gum formed.

The great variability in response to disease among the varieties of the same species and even in several areas of the trunk of the same individual suggest the desirability of a uniformly dependable biochemical test for the measurement of disease resistance. The senior author (*4*) has already indicated the possibility of using the degree of inhibition of the fungal enzymes by the bark of the several hosts as a test for disease resistance. It was reported that the bark of sour orange and of tangelo has consistently shown greater inhibition of the diastase of *Phytophthora citrophthora* of *Phomopsis californica* Fawcett, of saliva and malt than has that of lemon. Invertase of *Phytophthora citrophthora* similarly was inhibited more by sour orange than by lemon bark. Commercial invertase, however, was inhibited more by lemon than by sour orange. In all laboratory tests made subsequently lemon and sour orange have maintained their relative effects. Calamondin, however, which proved resistant in field tests, was about midway between sour orange and lemon in its degree of inhibition of fungous diastase. Another apparent failure of the parallelism was noted in a series in which the bark of a pummelo showed about the same inhibition of ptyalin as that of sour orange, but it should be recalled that some of the pummelos show resistance to the disease similar to that of sour orange. In general the laboratory tests approximated field observations; for example, one of the resistant citranges showed only slightly less effect on the fungous diastase than sour orange. Specimens of trifoliate orange, *Poncirus trifoliata*, in the Rubidoux plot, a species which has been used extensively as a stock in Japan and the Gulf States, proved more susceptible than lemon to the artificial inoculations, and in the laboratory less inhibitive than lemon.

While the results of laboratory tests with these carbohydrases taken as a whole are not striking they seem suggestive. It is not to be expected that all enzymes would be similarly affected. In

fact the enzyme emulsin was inhibited by sour orange only slightly more than by lemon, and urease action was stimulated by both. Likewise the pectinase of *Phomopsis californica* was inhibited only slightly and about equally by sour orange and lemon bark. This suggests that the failure of an organism to establish itself and progress in the tissues of its host may depend upon the destruction of or considerable inhibition of not necessarily all enzymes, but of one or more of its important enzymes by some cellular product of the host. Possibly the enzymes responsible for making carbon available are the important ones depressed or put out of action. In the case of a resistant host, such as sour orange, the paralyzing power may be thought of as being so great that the hyphae, handicapped by an insufficient carbon source due to the inhibition of their carbohydrases or other enzymes, can not progress so rapidly as the host callus forms; whereas in the susceptible host, as lemon, it may be postulated that there is a sufficient decrease in this antienzymic action to permit the hyphae to obtain food sufficiently rapidly for successful parasitism.

Application of the method to the proteases is the next step that suggests itself. The make-up of a species or variety is determined by the physical and chemical specificity of its protoplasm, and that in turn probably to a large extent by its own distinct proteinaceous materials. To account for the innumerable species and varieties one naturally turns to the chemistry of proteins. Of all biological substances the proteins only are sufficiently numerous to explain the specificity of such infinite variety. The infinite number of distinct proteins possible from combinations of 20 amino acids is strong support for the hypothesis that immunological specificity and probably most other features of biological specificity, as in heredity, must depend on these substances. The degree of disease production and establishment of a host-parasite relationship may depend largely on the ability of the microorganism to attack the food substances of the host, particularly the proteins.

While the extensive work reported here essentially substantiates the casual observations of many years, there are several apparent anomalies. These deviations from the expected show the necessity for making such field tests thorough and extensive. Before a species or variety is adopted commercially the response of a large number of individuals to the various pathogenes should be determined under each of the several possible sets of commercial conditions.

SUMMARY

A review of the literature is given, summarizing the range of susceptibility or resistance of the various species and varieties of Citrus and Citrus relatives to canker, West Indian lime anthracnose, withertip disease, scab, psorosis, decorticosis, and Pythiacystis gummosis.

The inoculation tests show that dependable comparisons of the response of species and varieties of Citrus to Pythiacystis gummosis can be made only on trees which are all of the same age and which are under similar environmental conditions.

Great variations in degree of susceptibility were shown by different varieties of the same species, individuals of the same variety, and

even by two or more locations on the same tree trunk, necessitating several inoculations on each tree to obtain a dependable average.

The varieties of the pummelo and citrange groups showed the largest differences in response to the disease, ranging from a susceptibility exceeding that in the lemon varieties to a resistance greater than the average of the sour orange group.

Under the conditions of the tests reported here the varieties of the individual groups as a whole rank in descending order of susceptibility as follows: Lemon, citrange, lime, pummelo and sweet orange, mandarin, rough lemon, tangelo, sour orange, and kumquat.

The amount of gum formed is approximately proportional to the severity and extent of the disease.

Calamondin (*Citrus mitis*), *C. hystrix*, *C. limetta*, *C. ichangensis*, Sampson tangelo, and the Cunningham, Sanford, and Savage varieties of citrange exhibited sufficient resistance to justify their trial as possible rootstocks.

The great variability found and the difficulties experienced in obtaining uniform trees and conditions for field tests suggest the desirability of dependable biochemical tests for resistance. Determination of the degree of inhibition of fungal enzymes by substances in the bark of the several hosts may be a practical laboratory procedure. Preliminary results of such biochemical tests and a comparison with field results are given.

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U. S. GOVERNMENT PRINTING OFFICE: 1930